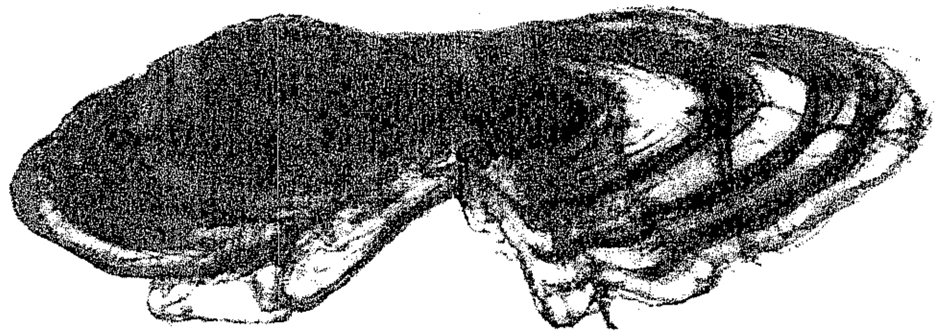
Final Results for 2004 Virginia - Chesapeake Bay Finfish Ageing with a Supplement of Tautog Ageing Research



by

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March 23, 2006

Final Summary Report

Development and Support of Facilities to Provide Finfish Ageing for Virginia Catches and Application of Virtual Population Analysis to Provide Management Advice

by

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Chapter 14 Comparison of Tautog Ageing

Executive Summary

In this report we present the results of ageing finfish collected from catches made in Virginia's marine waters in 2004. All fish were collected in 2004 by the Virginia Marine Resources Commission's (VMRC) Stock Assessment Program and aged in 2005 at the Center for Quantitative Fisheries Ecology's Age and Growth Laboratory at Old Dominion University. This report is broken down into chapters, one for each of the 13 species we aged. For each species, we present measures of ageing precision and bias, graphs of year-class distributions, and age-length keys.

For three species: summer flounder, *Paralichthys dentatus*, (n=380); striped bass, *Morone saxatilis*, (n=830); and tautog, *Tautoga onitis*, (n=506) multiple bony structures were used for determining fish age. Scales and otoliths were used to age summer flounder and striped bass, and opercula and otoliths were used to age tautog. Comparing alternative hard parts allowed us to assess their usefulness in determining fish age as well as the relative precision of each structure. Ages were determined from otoliths for the following species collected in Virginia waters during 2004: Atlantic croaker, *Micropogonias undulatus*, (n=331); black drum, *Pogonias cromis*, (n=16); bluefish, *Pomatomus saltatrix*, (n=326); cobia, *Rachycentron canadum*, (n=9); red drum, *Sciaenops ocellatus*, (n=6); spadefish, *Chaetodipterus faber*, (n=353); Spanish mackerel, *Scomberomorous maculates*, (n=430); spot, *Leiostomus xanthurus*, (n=459); spotted seatrout, *Cynoscion nebulosus*, (n=501); and weakfish, *Cynoscion regalis*, (n=657).

In total, we made 13,936 age readings from 6,168 scales, otoliths and opercula collected during 2004. A summary of the age ranges for all species aged is presented in Table I.

Species	Number of Fish	Number of Hard-Parts	Number of Age Readings	Minimum Age	Maximum Age
Atlantic croaker	331	331	762	2	13
black drum	16	16	132	2	4
bluefish	326	324	748	1	7
cobia	9	9	118	5	6
red drum	6	6	112	2	3
spadefish	353	351	802	1	20
Spanish mackerel	430	425	950	1	9
spot	459	458	1016	1	6
spotted seatrout	501	500	1100	0	3
striped bass	830	1400	3000	3	20
summer flounder	380	747	1694	0	10
tautog	506	944	2088	2	20
weakfish	657	657	1414	1	10
Totals	4804	6168	13936		

As part of our continued public outreach focused at recreational anglers, we again participated in the CCA's Kid's Fishing Day at Lynnhaven Fishing Pier. This was the fifth year our staff volunteered their time to participate in the event. We were also invited to a television show called the smoking gun outdoors hosted by Captain Chandler Hogg. During the show we explained our work and its importance in keeping Virginia fisheries healthy. We are proud to announce our Technician Roxanne Torres as well as our chief technician Eric Robillard received Portsmouth School Volunteer Service Award. Thanks to these two technicians, more than 150 students received a day of lessons. The training included age determination, morphology of different fish, habitat use, and importance of catch and release and following size limits. They also received hands on experience identifying fish organs and removing otoliths from fish donated from both commercial and recreational fisherman. We are currently working on a Bluefish *Species Update* report, which will be the fifth report in the series. Each report includes an overview of what is known about a fish species' biology, age and growth data and analyses generated in our lab, and interesting information on fish in general.

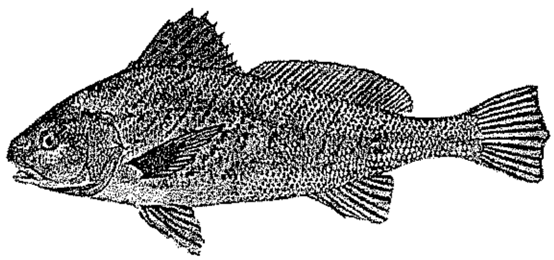
In 2004/2005 we upgraded our Age & Growth Laboratory website, which can be accessed at http://web.odu.edu/fish. The website includes electronic versions of this document along with more detailed explanations of the methods and structures we use in age determination.

Acknowledgements

We thank Roxanne Torres, Laura McCaskill, Mignonne Twine and Susan McKeel for their technical expertise in preparing otoliths, scales, and opercula for age determination. Roxanne, Susan, and Serena Turner all put in long hours processing "tons" of fish in our lab. We are also thankful for Dr. William Persons' III hard work on our *Species Updates* and web page. A special note of appreciation to Ron Owens, Joanie Beatley, and Myra Thompson for their many efforts in this cooperative project. This work was funded by the Virginia Saltwater Recreational Development Fund.

The image on the front cover is an otolith thin-section from a 415 mm (16.3 inch) total length, 4 year-old male tautog. The fourth annulus is forming at the edge of the otolith.

Chapter 1
Atlantic Croaker



Micropogonias undulatus

INTRODUCTION

A total of 331 Atlantic croaker, *Micropogonias undulatus*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age was 6.2 years, and the standard deviation and standard error were 2.3 and 0.1, respectively. Twelve age classes (2 to 13) were represented, comprising fish from the 1991-2002 year-classes. Fish from the 1997 through 2001 year-classes dominated the sample

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. In the lab they were sorted by date of capture, their envelope labels were verified against VMRC's collection data, and each fish was assigned a unique Age and Growth Laboratory identification number. All otoliths were stored dry in labeled cell well plates.

Preparation — Otoliths were processed following the methods described in Barbieri et al. (1994) with a few modifications. Briefly, the left or right sagittal otolith was randomly selected and attached to a glass slide with Aremco's clear Crystalbond™ 509 adhesive. At least two serial transverse sections were cut through the core of each otolith with a Buehler Isomet low-speed saw equipped with a three inch, fine grit Norton diamond-wafering blade. Otolith sections were placed on labeled glass slides and covered with a thin layer of Flo-texx mounting medium, that not only adhered the sections to the slide, but more importantly, provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — Sectioned otoliths were aged by two different readers using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 20 times magnification. Each reader aged all of the otolith samples. The ageing criteria reported in Barbieri et al. (1994) were used in age determination, particularly regarding the location of the first annulus (Figure 1).

All samples were aged in chronological order, based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

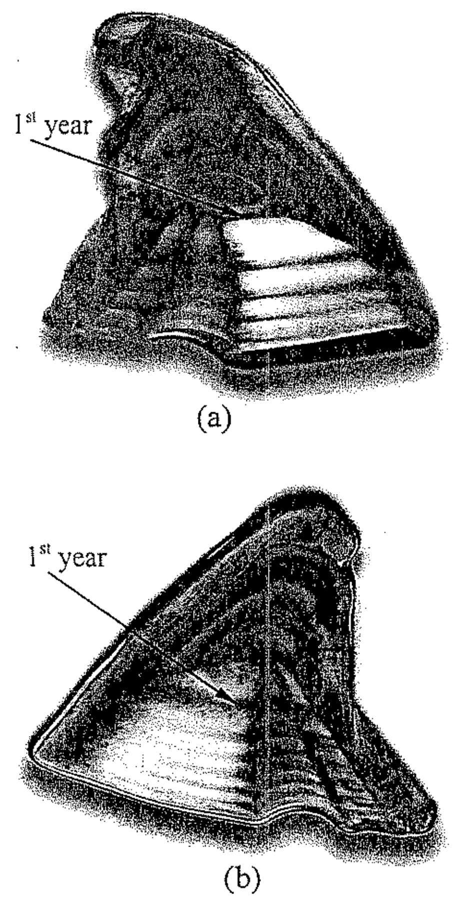


Figure 1. Otolith cross-sections of a) a 5 year old croaker with a small 1st annulus, and b) a 6 year old croaker with a large 1st annulus.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. To detect any changes or drift in our ageing methods, both readers re-aged the otoliths of 50 randomly selected fish previously aged in 2000. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

No bias was discovered in any of the self-precision tests of otolith age estimates, with both readers equally able to reproduce the ages of previously read samples. There was also 99.1 percent agreement between reader age estimates. Figure 2 illustrates the between readers' precision of age estimates.

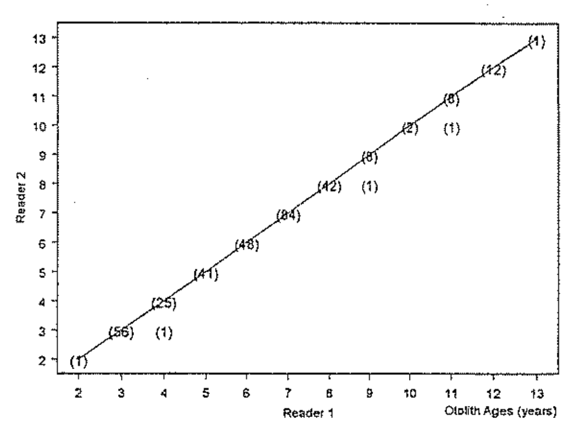


Figure 2. Between-reader comparison of otolith age estimates for Atlantic croaker.

Of the 331 fish aged with otoliths, 12 age classes (2 to 13) were represented (Table 1). The average age for the sample was 6.2 years, and the standard deviation and standard error were 2.3 and 0.1, respectively.

Year-class data (Figure 3) indicate that recruitment into the fishery begins at age 2, but large numbers are not seen until age 3, which corresponds to the 2001 year-class for Atlantic croaker collected in 2004. While the ratio of males to females shows an overall higher number of females, both sexes show trends of high abundance for the 1996 through 2001 year-classes.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

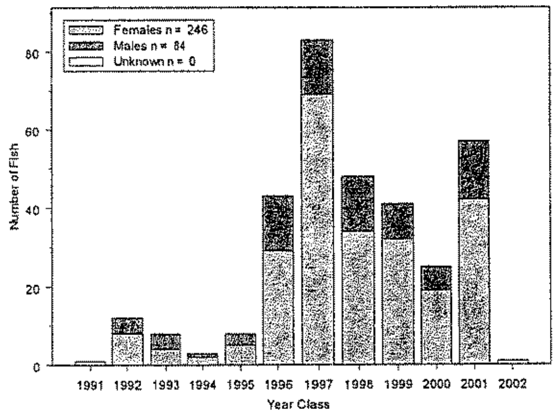


Figure 3. Year-class distribution for Atlantic croaker collected for ageing in 2004. Distributions are broken down by sex.

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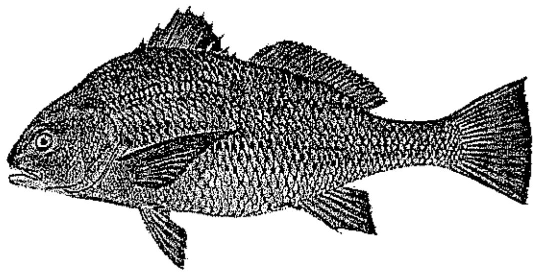
Table 1. The number of Atlantic croaker assigned to each total length-at-age category for 331 fish sampled for age determination in Virginia during 2004. Length not reported for 1 fish.

Length 1-inch intervals	Age (years)												
	2	3	4	5	6	7	8	9	10	11	12	13	Totals
8 - 8.99	1	0	0	0	0	0	0	0	0	0	0	0	1
9 - 9.99	0	3	0	0	0	0	0	0	0	0	0	0	3
10 - 10.99	0	9	3	0	0	0	0	0	0	0	0	0	12
11 - 11.99	0	29	5	11	6	4	0	0	0	0	0	0	55
12 - 12.99	0	15	15	19	14	21	6	1	0	1	0	0	92
13 - 13.99	0	1	2	6	14	19	5	0	0	0	0	0	47
14 - 14.99	0	0	0	2	5	14	10	1	0	0	1	0	33
15 - 15.99	0	0	0	0	5	12	12	2	2	4	2	0	39
16 - 16.99	0	0	0	1	2	8	6	1	1	2	3	1	25
17 - 17.99	0	0	0	1	2	5	1	3	0	1	3	0	16
18 - 18.99	0	0	0	0	0	1	3	0	0	0	0	0	4
19 - 19.99	0	0	0	0	0	0	0	0	0	0	2	0	2
20 - 20.99	0	0	0	0	0	0	0	0	0	0	1	0	1
Totals	1	57	25	40	48	84	43	8	3	8	12	1	330

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for Atlantic croaker sampled for age determination in Virginia during 2004. Length not reported for 1 fish

Length 1-inch intervals	Age (years)												
	2	3	4	5	6	7	8	9	10	11	12	13	N
8 - 8.99	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1
9 - 9.99	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	3
10 - 10.99	0.000	0.750	0.250	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	12
11 - 11.99	0.000	0.527	0.091	0.200	0.109	0.073	0.000	0.000	0.000	0.000	0.000	0.000	55
12 - 12.99	0.000	0.163	0.163	0.207	0.152	0.228	0.065	0.011	0.000	0.011	0.000	0.000	92
13 - 13.99	0.000	0.021	0.043	0.128	0.298	0.404	0.106	0.000	0.000	0.000	0.000	0.000	47
14 - 14.99	0.000	0.000	0.000	0.061	0.152	0.424	0.303	0.030	0.000	0.000	0.030	0.000	33
15 - 15.99	0.000	0.000	0.000	0.000	0.128	0.308	0.308	0.051	0.051	0.103	0.051	0.000	39
16 - 16.99	0.000	0.000	0.000	0.040	0.080	0.320	0.240	0.040	0.040	0.080	0.120	0.040	25
17 - 17.99	0.000	0.000	0.000	0.063	0.125	0.313	0.063	0.188	0.000	0.063	0.188	0.000	16
18 - 18.99	0.000	0.000	0.000	0.000	0.000	0.250	0.750	0.000	0.000	0.000	0.000	0.000	4
19 - 19.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	2
20 - 20.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	1
											Sample Size		330

Chapter 2
Black Drum



Pogonias cromis

INTRODUCTION

A total of 16 black drum, *Pogonias cromis*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age of the sample was 3.0 years, with a standard deviation of 0.44 and a standard error of 0.11. The youngest fish was a two year old and the oldest fish was 4 years old, representing the 2000 and 2002 year-classes, respectively.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. In the lab they were sorted by date of capture, their envelope labels were verified against VMRC's collection data, and each fish was assigned a unique Age and Growth Laboratory sample number. All otoliths were stored dry in their original VMRC coin envelopes.

Preparation — Otoliths were processed for ageing following the methods described in Bobko (1991) and Jones and Wells (1998). Briefly, at least two serial transverse sections were cut through the nucleus of each otolith with a Buehler Isomet low-speed saw equipped with a three inch, fine grit Norton diamond-wafering blade. Otolith sections were placed on labeled glass slides and covered with a thin layer of Flo-texx mounting medium, that not only adhered the sections to the slide, but more importantly, provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — Sectioned otoliths were aged by two different readers using a Leica MZ-12 dissecting microscope with transmitted light at between 8 and 20 times magnification (Figure 1). Each reader aged all of the otolith samples.

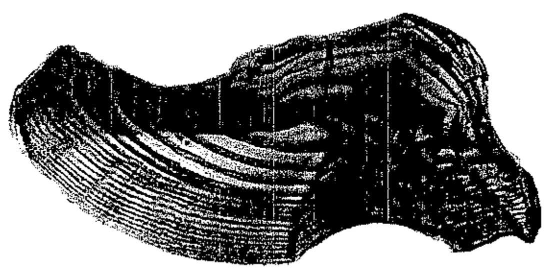


Figure 1. Otolith thin-section from a 20 year-old black drum.

All samples were aged in chronological order, based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, both readers aged all fish a second time to measure reader precision and age reproducibility. To detect any changes or drift in our ageing methods, both readers re-aged the otoliths of 50 randomly selected fish previously aged in 2000. We considered a reader to be biased if the readings revealed consistent over or under ageing. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

No bias was discovered in any of the self-precision tests of otolith age estimates, with both readers equally able to reproduce the ages of previously read samples. There was also 88 percent agreement between reader age estimates. Figure 2 illustrates the

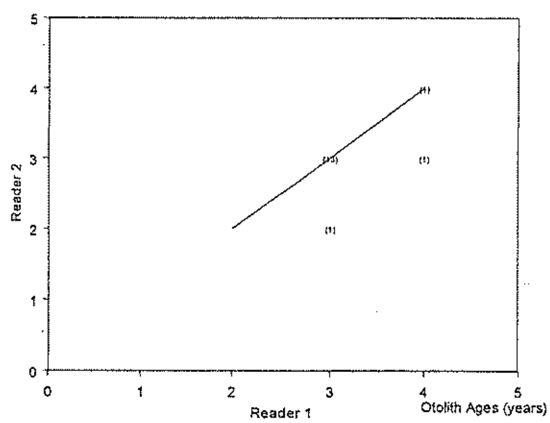


Figure 2. Between-reader comparison of otoliths age estimates for black drum.

between readers' precision of age estimates.

Of the 16 fish aged with otoliths, 3 age classes were represented (Table 1). The average age of the sample was 3.0 years, with a standard deviation of 0.44 and a standard error of 0.11. The youngest fish was a two year old and the oldest fish was 4 years old, representing the 2002 and 2000 year-classes, respectively. Year-class data (Figure 3) show that the sample was comprised of 3 year-classes, comprising fish from the 2000, 2001 and 2002 year-classes, with fish primarily from the 2001 year-class.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

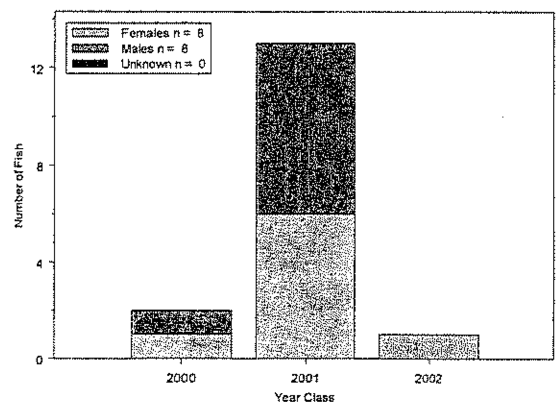


Figure 3. Year-class distribution for black drum collected for ageing in 2003. Distributions are broken down by sex.

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Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

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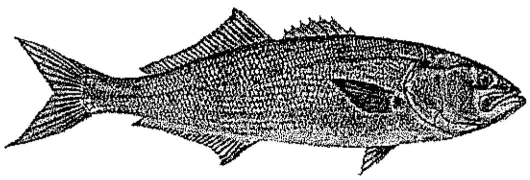
Table 1. The number of black drum assigned to each total length-at-age category for 16 fish sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)			
	2	3	4	Totals
19 - 19.99	0	1	0	1
20 - 20.99	0	1	0	1
21 - 21.99	0	1	0	1
22 - 22.99	0	5	0	5
23 - 23.99	1	3	0	4
24 - 24.99	0	2	1	3
26 - 26.99	0	0	1	1
Totals	1	13	2	16

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for black drum sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)			
	2	3	4	N
19 - 19.99	0.00	1.00	0.00	1
20 - 20.99	0.00	1.00	0.00	1
21 - 21.99	0.00	1.00	0.00	1
22 - 22.99	0.00	1.00	0.00	5
23 - 23.99	0.25	0.75	0.00	4
24 - 24.99	0.00	0.67	0.33	3
26 - 26.99	0.00	0.00	1.00	1
			Samples Size	16

Chapter 3
Bluefish



Pomatomus saltatrix

INTRODUCTION

A total of 326 bluefish, *Pomatomus saltatrix*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. We were unable to age two fish due to the poor quality of their otoliths. The average age for the 324 aged fish was 1.7 years, and the standard deviation and standard error were 0.8 and 0.04, respectively. Six age classes (1 to 3 and 5 to 7) were represented, comprising fish from the 1997-1999 and 2001 to 2003 year-classes. The 2002 and 2003 year-classes dominated the sample.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and assigned unique Age and Growth Laboratory sample numbers. All otoliths were stored dry in labeled cell well plates.

Preparation — We used a bake and thin-section technique to process bluefish otoliths for age determination. Otolith preparation began by randomly selecting either the right or left otolith. Each otolith was mounted with Crystal Bond onto a standard microscope slide with its distal surface orientated upwards. Once mounted, a small mark was placed on the otolith surface directly above the otolith focus. The slide, with attached otolith, was then secured to an Isomet saw equipped with two diamond wafering blades separated by a 0.5 mm spacer, which was slightly smaller in diameter than the diamond blades. The otolith was positioned so that the wafering blades straddled each side of the otolith focus ink mark. It was crucial that this cut be perpendicular to the long axis of the otolith. Failure to do so resulted in "broadening" and distortion of winter growth zones. A proper cut resulted in annuli that were clearly defined and delineated. Once cut, the otolith section was placed into a ceramic "Coors" spot plate well and baked in a Thermolyne 1400 furnace at 400°C. Baking time was otolith size dependent and gauged by color, with a light caramel color desired. Once a suitable color was reached the baked thin-section was placed on a labeled glass slide and covered with a thin layer of Flo-texx mounting medium, that not only adhered the sections to the slide, but more importantly, provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — Two different readers using a LEICA MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification aged all sectioned otoliths (Figure 1). If an otolith was properly sectioned the sulcal groove came to a sharp point within the middle of the focus. Typically the first year's annulus was found by locating the focus of the otolith, which was characterized as a visually distinct dark oblong region found in the center of the otolith. The first year's annulus had the highest visibility proximal to the focus along the edge of the sulcal groove. Once located, the first year's annulus was followed outward from the sulcal groove towards the dorsal perimeter of the otolith. Often, but not always, the first year was associated with a very distinct crenellation on the dorsal surface and a prominent protrusion on the ventral surface. Unfortunately both these landmarks had a tendency to become less prominent in older fish.

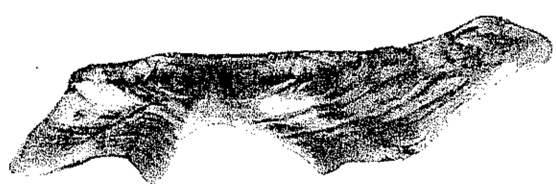


Figure 1. Otolith thin-section from a 850mm TL 8 year-old female bluefish.

Even with the bake and thin-section technique, interpretation of the growth zones from the otoliths of young bluefish was difficult. Rapid growth within the first year of life prevents a sharp delineation between opaque and translucent zones. When the exact location of the first year was not clearly evident, and the otolith had been sectioned accurately, a combination of surface landscape (1st year crenellation) and the position of the second annuli were used to help determine the position of the first annulus.

What appeared to be "double annuli" were occasionally observed in bluefish four years of age and older. This annulus formation typically occurred within years 4 to 7, and was characterized by distinct and separate annuli in extremely close proximity to each other. We do not know if the formation of these double annuli were two separate annuli, or in fact only one, but they seemed to occur during times of reduced growth after maturation. "Double annuli" were considered to be one annulus when both marks joined to form a central origin. The origins being the sulcal groove and at the outer peripheral edge of the otolith. If these annuli did not meet to form a central origin they were considered two annuli, and counted as such.

All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

The measurement of reader self-precision was not high for both readers (reader 1's

CV = 14.2% and reader 2's CV = 1.69%). There was evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 16.5$, df = 3, $P = 0.0008$). Figure 2 illustrates the between readers' precision of age estimates. The average coefficient of variation (CV) of 8.6% was significant.

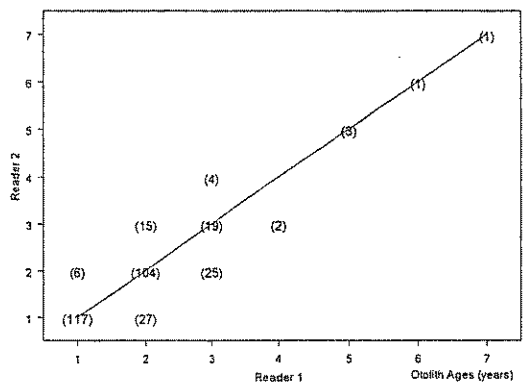


Figure 2. Between-reader comparison of otolith age estimates for bluefish.

Of the 326 fish aged with otoliths 6 age classes were represented (Table 1). The average age for the sample was 1.67 years, and the standard deviation and standard error were 0.8 and 0.04, respectively.

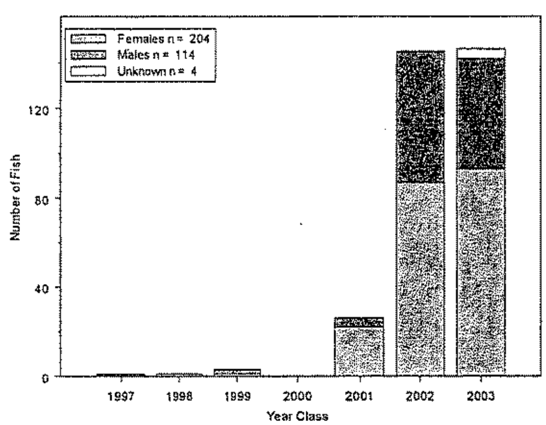


Figure 3. Year-class distribution for bluefish collected for ageing in 2004. Distribution is broken down by sex.

Year-class data (Figure 3) indicates that recruitment into the fishery began at age 1, which corresponded to the 2003 year-class for bluefish caught in 2004. One and Two-year-old fish were the dominant year-class in the 2004 sample.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

REFERENCES

Campana, S.E., M.C. Annand, and J.I. McMillan. 1995. Graphical and statistical methods for determining the consistency of age determinations. Trans. Am. Fish. Soc. 124:131-138.

Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

S-Plus. 1999. S-Plus 4.5 Guide to Statistics. Data Analysis Products Division. Math Soft, Inc. Seattle, Washington.

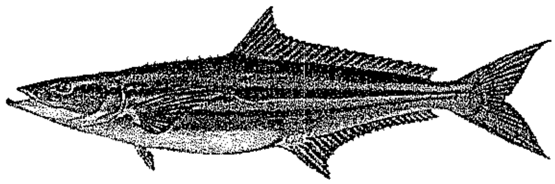
Table 1. The number of bluefish assigned to each total length-at-age category for 326 fish collected for age determination in Virginia in 2004 (length not reported for 3 fish).

Length 1-inch intervals	Age (years)						
	1	2	3	5	6	7	Totals
7 - 7.99	2	0	0	0	0	0	2
8 - 8.99	1	0	0	0	0	0	1
9 - 9.99	12	0	0	0	0	0	12
10 - 10.99	21	0	0	0	0	0	21
11 - 11.99	23	0	0	0	0	0	23
12 - 12.99	33	0	0	0	0	0	33
13 - 13.99	24	0	0	0	0	0	24
14 - 14.99	18	3	0	0	0	0	21
15 - 15.99	9	8	0	0	0	0	17
16 - 16.99	4	22	3	0	0	0	29
17 - 17.99	1	31	7	0	0	0	39
18 - 18.99	0	51	6	0	0	0	57
19 - 19.99	0	22	5	0	0	0	27
20 - 20.99	0	7	2	0	0	0	9
21 - 21.99	0	1	0	0	0	0	1
22 - 22.99	0	0	1	0	0	0	1
23 - 23.99	0	0	1	0	0	0	1
24 - 24.99	0	0	1	0	0	0	1
27 - 27.99	0	0	0	1	0	0	1
29 - 29.99	0	0	0	1	1	0	2
31 - 31.99	0	0	0	1	0	0	1
Totals	148	145	26	3	1	0	323

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages, for bluefish collected for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years) 1	2	3	5	6	7	N
7 - 7.99	1.000	0.000	0.000	0.000	0.000	0.000	2
8 - 8.99	1.000	0.000	0.000	0.000	0.000	0.000	1
9 - 9.99	1.000	0.000	0.000	0.000	0.000	0.000	12
10 - 10.99	1.000	0.000	0.000	0.000	0.000	0.000	21
11 - 11.99	1.000	0.000	0.000	0.000	0.000	0.000	23
12 - 12.99	1.000	0.000	0.000	0.000	0.000	0.000	33
13 - 13.99	1.000	0.000	0.000	0.000	0.000	0.000	24
14 - 14.99	0.857	0.143	0.000	0.000	0.000	0.000	21
15 - 15.99	0.529	0.471	0.000	0.000	0.000	0.000	17
16 - 16.99	0.138	0.759	0.103	0.000	0.000	0.000	29
17 - 17.99	0.026	0.795	0.179	0.000	0.000	0.000	39
18 - 18.99	0.000	0.895	0.105	0.000	0.000	0.000	57
19 - 19.99	0.000	0.815	0.185	0.000	0.000	0.000	27
20 - 20.99	0.000	0.778	0.222	0.000	0.000	0.000	9
21 - 21.99	0.000	1.000	0.000	0.000	0.000	0.000	1
22 - 22.99	0.000	0.000	1.000	0.000	0.000	0.000	1
23 - 23.99	0.000	0.000	1.000	0.000	0.000	0.000	1
24 - 24.99	0.000	0.000	1.000	0.000	0.000	0.000	1
27 - 27.99	0.000	0.000	0.000	1.000	0.000	0.000	1
29 - 29.99	0.000	0.000	0.000	0.500	0.500	0.000	2
31 - 31.99	0.000	0.000	0.000	1.000	0.000	0.000	1
					Sample Size		323

Chapter 4
Cobia



Rachycentron canadum

INTRODUCTION

A total of 9 cobia, *Rachycentron canadum*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age of the sample was 5.5 years, and the standard deviation and standard error were 0.52 and 0.17, respectively. Two age classes (5 and 6) were represented, comprising fish from the 1998 and 1999 year-classes. The 1998 year-class dominated the sample.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and assigned unique Age and Growth Laboratory sample numbers. All otoliths were stored dry in labeled cell well plates.

Preparation — Individual otoliths were placed into 14 mm x 5 mm x 3 mm wells (Ladd Industries silicon rubber mold) filled with Loctite adhesive. Each otolith was rolled around in the Loctite to remove all trapped air bubbles and ensure complete coverage of the otolith surface. The otoliths were oriented sulcal side down with the long axis of the otolith exactly parallel with the long axis of the mold well. Once the otoliths were properly oriented, the mold was placed under UV light and left to solidify overnight. Once dry, each embedded otolith was removed from the mold and mounted with Crystal Bond onto a standard microscope slide. Once mounted, a small mark was placed on the otolith surface directly above the otolith focus. The slide, with attached otolith, was then secured to an Isomet saw equipped with two diamond wafering blades separated by a 0.5 mm spacer, which was slightly smaller in diameter than the diamond blades. The otolith was positioned so that the wafering blades straddled each side of the focus ink mark. The glass slide was adjusted to ensure that the blades were exactly perpendicular to the long axis of the otolith. The otolith wafer section was viewed under a dissecting microscope to determine which side (cut surface) of the otolith was closer to the focus. The otolith section was mounted best-side up onto a glass slide with Crystal Bond. The section was then lightly polished on a Buehler Ecomet 3 variable speed grinder-polisher with Mark V Laboratory 30-micron polishing film. After drying, a thin layer of Flo-texx mounting medium was applied over the polished otolith surface, which provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — Two different readers using a LEICA MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification aged all sectioned otoliths (Figure 1). Both age readers aged all of the otolith samples.

All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

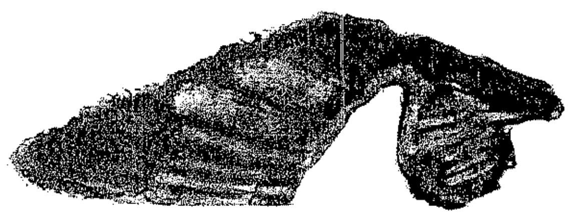


Figure 1. Otolith thin-section from a 1524mm TL 6 year old cobia.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, both readers aged all fish a second time to measure reader precision and age reproducibility. To detect any changes or drift in our ageing methods, both readers re-aged the otoliths of 50 randomly selected fish previously aged in 2000. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

No bias was discovered in any of the self-precision tests of otolith age estimates, with both readers equally able to reproduce the ages of previously read samples. There was also 90 percent agreement between reader age estimates. Figure 2 illustrates the between readers' precision of age estimates.

Of the 9 fish aged, 2 age classes were represented (Table 1). The average age of the sample was 5.5 years, and the standard deviation and standard error were 0.52 and 0.17, respectively.

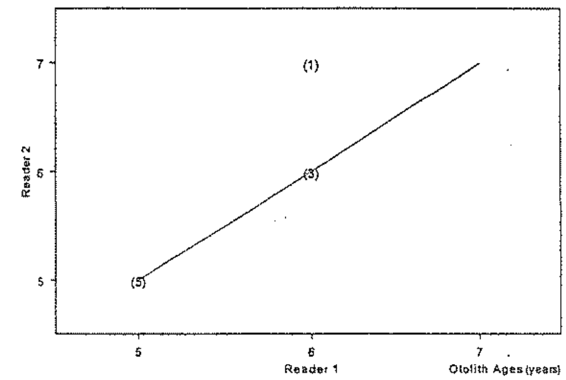


Figure 2. Between-reader comparison of otolith age estimates for cobia.

Year-class data (Figure 3) indicates that recruitment into the fishery begins at age 5, which corresponds to the 1999 year-class for cobia caught in 2004. The year-class 1998 dominated the sample.

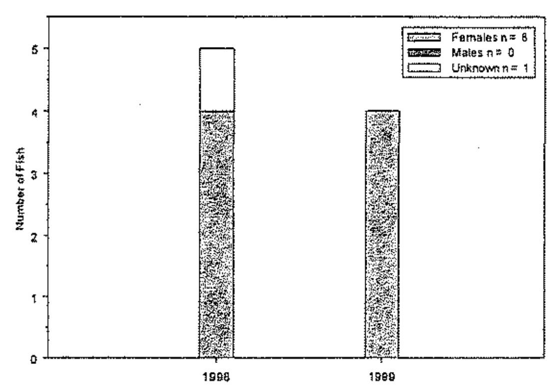


Figure 3. Year-class distribution for cobia collected for ageing in 2004. Distribution is broken down by sex.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

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Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

S-Plus. 1999. S-Plus 4.5 Guide to Statistics. Data Analysis Products Division. Math Soft, Inc. Seattle, Washington.

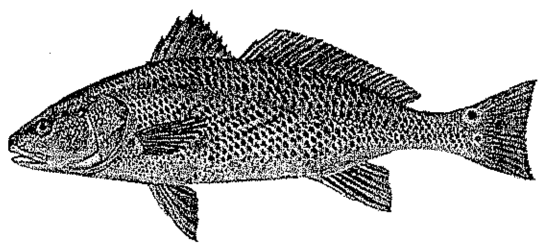
Table 1. The number of cobia assigned to each total length-at-age category for 9 fish sampled for age determinatior in Virginia during 2004.

Length 1-inch intervals	Age (years)		
	5	6	Totals
42 - 42.99	0	1	1
46 - 46.99	1	0	1
49 - 49.99	1	0	1
50 - 50.99	1	0	1
51 - 51.99	0	1	1
52 - 52.99	0	1	1
53 - 53.99	1	1	2
54 - 54.99	0	1	1
Totals	4	5	9

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for cobia sampled for age determination in Virginia during 2004.

Length 1-inch intervals	5	6	N
42 - 42.99	0.000	1.000	1
46 - 46.99	1.000	0.000	1
49 - 49.99	1.000	0.000	1
50 - 50.99	1.000	0.000	1
51 - 51.99	0.000	1.000	1
52 - 52.99	0.000	1.000	1
53 - 53.99	0.500	0.500	2
54 - 54.99	0.000	1.000	1
		Sample Size	9

Chapter 5
Red Drum
Sciaenops ocellatus



INTRODUCTION

A total of 6 red drum, *Sciaenops ocellatus*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age of the sample was 2.8 years, and the standard deviation and standard error were 0.4 and 0.16, respectively. Two age classes (2 and 3) were represented, comprising fish from the 2001-2002 year-classes. All of the fish were three years-of-age or less.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and assigned unique Age and Growth Laboratory sample numbers. All otoliths were stored dry in their original labeled coin envelopes.

Preparation — Otoliths were processed for ageing following the methods described in Bobko (1991) for black drum. Briefly, otoliths were mounted on glass slides with Crystal Bond. At least two serial transverse sections were cut through the nucleus of each otolith with a Buehler Isomet low-speed saw equipped with a three inch, fine grit Norton diamond-wafering blade. After drying, a thin layer of Flo-texx mounting medium was applied to the otolith section to increase light transmission through the translucent zones, which provided enhanced contrast and greater readability.

Readings — Two different readers aged all sectioned otoliths using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 20 times magnification (Figure 1).

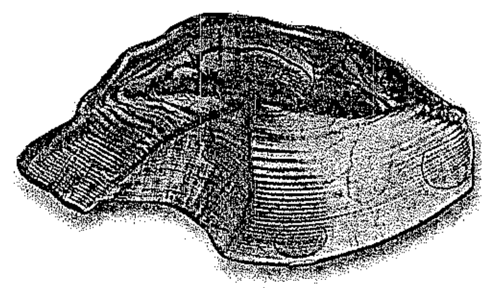


Figure 1. Otolith thin-section from 26 year old red drum.

All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Red drum ages were based on a biological birthdate of September 1, while year-class assignment was based on a January 1 annual birthdate. Red drum were treated in this manner because of the timing of spawning and the fact that the first annulus is not seen on an otolith until a fish's second spring. For example, a red drum that was born in September of 1997 and captured in March of 1999 would not have any visible annuli on it's otoliths, but would be aged as a 1 year-old fish since it lived beyond one September (September 1998). But this 1 year-old fish caught in 1999 would be mistakenly assigned to the 1998 year-class. In order to properly assign the fish to its correct year-class, 1997, a January birthdate was used which would make the fish 2 years-old (since the fish lived past January 1998 and 1999) and year-class would be assigned correctly.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, both readers aged all 6 fish a second time to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

Measurements of reader self-precision were high, with both readers able to reproduce 100 % of the ages of previously read otoliths. Figure 2 illustrates the between readers' precision of age estimates. There was 85% agreement between readers.

Of the 6 fish aged with otoliths, 2 age classes were represented (Table 1). The average age of the sample was 2.8 years, and the standard deviation and standard error were 0.4 and 0.16, respectively.

Year-class data (Figure 3) indicate that the 2001 year-class dominated the sample. Indicative of the trend in the recreational fishing, very few older fish were collected in 2004.

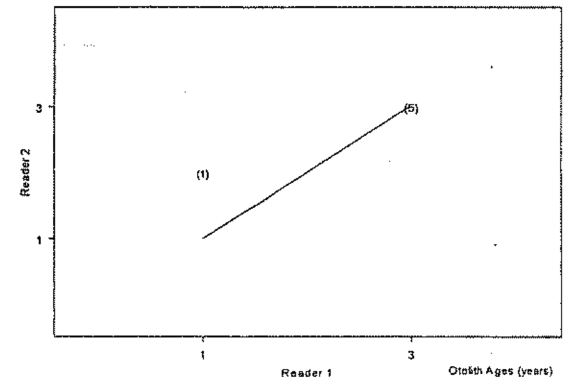


Figure 2. Between-reader comparison of otolith age estimates for red drum

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

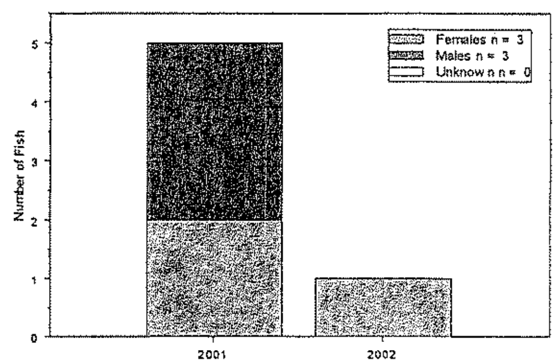


Figure 3. Year-class distribution for red drum collected for ageing in 2004. Distribution is broken down by sex.

REFERENCES

Bobko, S. J. 1991. Age, growth, and reproduction of black drum, *Pogonias cromis*, in Virginia. M.S. thesis. Old Dominion University, Norfolk, VA.

Campana, S.E., M.C. Annand, and J.I. McMillan. 1995. Graphical and statistical methods for determining the consistency of age determinations. Trans. Am. Fish. Soc. 124:131-138.

Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

S-Plus. 1999. S-Plus 4.5 Guide to Statistics. Data Analysis Products Division. Math Soft, Inc. Seattle, Washington.

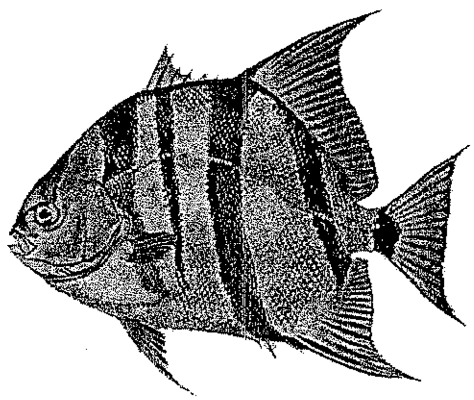
Table 1. The number of red drum assigned to each total length-at-age category for 6 fish sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)		
	2	3	Totals
18 - 18.99	1	0	1
24 - 24.99	0	2	2
25 - 25.99	0	3	3
Totals	1	5	6

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for red drum sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)		
	2	3	N
18 - 18.99	1.000	0.000	1
24 - 24.99	0.000	1.000	2
25 - 25.99	0.000	1.000	3
		Sample Size	6

Chapter 6
Atlantic Spadefish



Chaetodipterus faber

INTRODUCTION

A total of 353 spadefish, *Chaetodipterus faber*, was collected for age and growth analysis in 2004. We were unable to age two fish due to poor quality of their otoliths. The average age of the sample was 2.7 years, and the standard deviation and standard error were 2.4 and 0.1, respectively. Sixteen age classes (1 to 3 and 5 to 16 and 20) were represented, comprising fish from the 1984 and 1988-1999 and 2001-2003 year-classes.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and assigned unique Age and Growth Laboratory sample numbers. All otoliths were stored dry in labeled cell well trays.

Preparation — Otoliths were processed for ageing using a bake and thin-section technique. Preparation began by randomly selecting either the right or left otolith. The otolith was mounted with Crystal Bond onto a standard microscope slide with its distal surface orientated upwards. Once mounted, a small mark was placed on the otolith surface directly above the otolith focus. The slide, with attached otolith, was then secured to a Buehler Isomet low-speed saw equipped with two fine grit Norton diamond-wafering blades separated by a 0.5 mm spacer, which was slightly smaller in diameter than the diamond blades. The otolith was positioned so that the wafering blades straddled each side of the otolith focus ink mark. It was crucial that this cut be perpendicular to the long axis of the otolith. Failure to do so resulted in "broadening" and distortion of winter growth zones. A proper cut resulted in annuli that were clearly defined and delineated. Once cut, the otolith section was placed into a ceramic "Coors" spot plate well and baked in a Thermolyne 1400 furnace at 400°C. Baking time was otolith size dependent and gauged by color, with a light caramel color desired. Once a suitable color was reached the baked thin-section was placed on a labeled glass slide and covered with a thin layer of Flo-texx mounting medium, which provided enhanced contrast and greater readability by increasing light transmission through the sections.

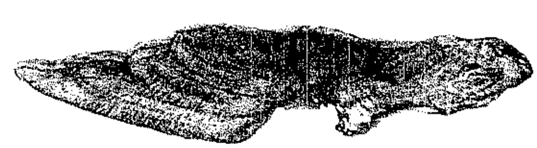


Figure 1. Sectioned otolith from a 3-year-old female spadefish.

Readings — Two different readers aged all sectioned otoliths using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 1).

All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

Measurements of reader self-precision were high, with both readers able to reproduce the ages of previously read otoliths (reader 1's CV = 5.0% and reader 2's CV = 2.2%). Figure 2 illustrates the between readers' precision of age estimates, with only four age difference greater than one year. There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 20$, df = 13, $P = 0.09$). The average coefficient of variation (CV) of 5% was considered not to be significant and lower than the CV of 5.6% reported in 2002.

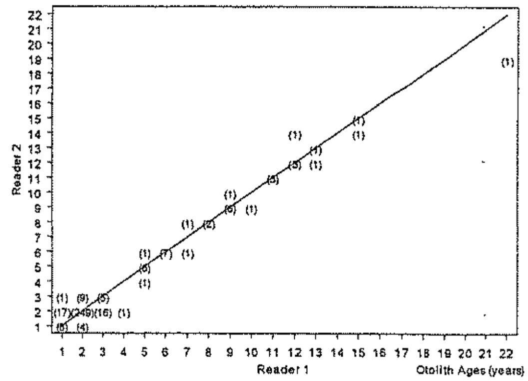


Figure 2. Between-reader comparison of otolith age estimates for spadefish.

Of the 351 fish aged with otoliths, 16 age classes were represented (Table 1). The average age of the sample was 2.7 years, and the standard deviation and standard error were 2.4 and 0.1, respectively. Year-class data (Figure 3) indicate that the 2002 year-class dominated the sample.

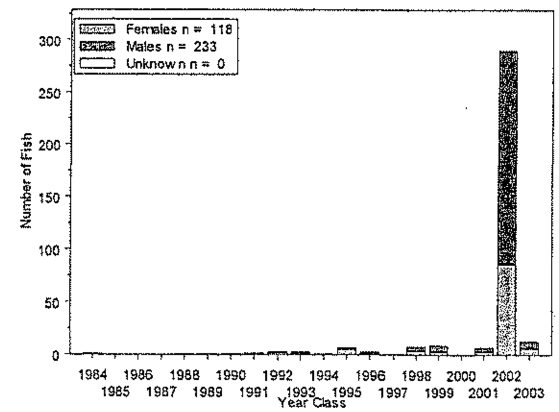


Figure 3. Year-class distribution for spadefish collected for ageing in 2004. Distribution is broken down by sex.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

REFERENCES

Campana, S.E., M.C. Annand, and J.I. McMillan. 1995. Graphical and statistical methods for determining the consistency of age determinations. Trans. Am. Fish. Soc. 124:131-138.

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S-Plus. 1999. S-Plus 4.5 Guide to Statistics. Data Analysis Products Division. Math Soft, Inc. Seattle, Washington.

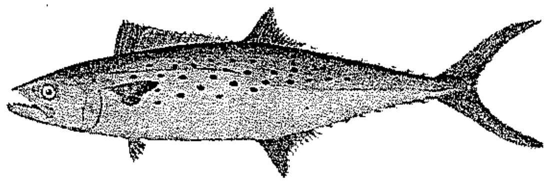
Table 1. The number of spadefish assigned to each total length-at-age category for 353 fish collected for age determination in Virginia during 2004. Length not reported for 3 fish.

Length 1-inch intervals	Age (years)																
	1	2	3	5	6	7	8	9	10	11	12	13	14	15	16	20	Totals
5 - 5.99	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
6 - 6.99	5	33	0	0	0	0	0	0	0	0	0	0	0	0	0	0	38
7 - 7.99	4	103	0	0	0	0	0	0	0	0	0	0	0	0	0	0	107
8 - 8.99	1	80	0	0	0	0	0	0	0	0	0	0	0	0	0	0	81
9 - 9.99	2	53	0	0	0	0	0	0	0	0	0	0	0	0	0	0	55
10 - 10.99	0	13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	13
11 - 11.99	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5
12 - 12.99	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
14 - 14.99	0	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	2
15 - 15.99	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	3
16 - 16.99	0	0	1	1	0	0	0	0	0	0	0	0	0	0	0	0	2
17 - 17.99	0	0	2	4	1	0	0	0	0	0	0	0	0	0	0	0	7
18 - 18.99	0	0	0	2	3	0	0	1	0	0	0	0	0	0	0	0	6
19 - 19.99	0	0	0	2	0	0	3	1	0	1	0	0	0	0	0	0	7
20 - 20.99	0	0	0	0	2	0	0	3	1	0	2	1	0	0	0	0	9
21 - 21.99	0	0	0	0	1	0	0	1	0	1	1	1	0	1	0	0	6
22 - 22.99	0	0	0	0	0	1	0	1	0	1	0	0	1	0	1	0	5
24 - 24.99	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1
Totals	13	290	7	9	7	1	3	7	1	3	3	2	1	1	1	1	350

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for spadefish sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)																
	1	2	3	5	6	7	8	9	10	11	12	13	14	15	16	20	N
5 - 5.99	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1
6 - 6.99	0.13	0.87	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	38
7 - 7.99	0.04	0.96	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	107
8 - 8.99	0.01	0.99	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	81
9 - 9.99	0.04	0.96	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	55
10 - 10.99	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	13
11 - 11.99	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	5
12 - 12.99	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	2
14 - 14.99	0.00	0.50	0.50	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	2
15 - 15.99	0.00	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	3
16 - 16.99	0.00	0.00	0.50	0.50	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	2
17 - 17.99	0.00	0.00	0.29	0.57	0.14	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	7
18 - 18.99	0.00	0.00	0.00	0.33	0.50	0.00	0.00	0.17	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	6
19 - 19.99	0.00	0.00	0.00	0.29	0.00	0.00	0.43	0.14	0.00	0.14	0.00	0.00	0.00	0.00	0.00	0.00	7
20 - 20.99	0.00	0.00	0.00	0.00	0.22	0.00	0.00	0.33	0.11	0.00	0.22	0.11	0.00	0.00	0.00	0.00	9
21 - 21.99	0.00	0.00	0.00	0.00	0.17	0.00	0.00	0.17	0.00	0.17	0.17	0.17	0.00	0.17	0.00	0.00	6
22 - 22.99	0.00	0.00	0.00	0.00	0.00	0.20	0.00	0.20	0.00	0.20	0.00	0.00	0.20	0.00	0.20	0.00	5
24 - 24.99	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.00	1
															Sample Size		350

Chapter 7
Spanish Mackerel



Scomberomorous maculatus

INTRODUCTION

A total of 430 Spanish mackerel, *Scomberomorous maculatus*, was collected by the Virginia Marine Resource Commission (VMRC) Stock Assessment Program and the Center for Quantitative Fisheries Ecology (CQFE). Age was determined for 425 Spanish mackerel using sagittal otoliths. The average age for the 425 fish was 1.8 years, and the standard deviation and standard error were 1.1 and 0.05, respectively. Seven age classes were observed (1 – 6 and 9), representing fish from the 1998 through 2003 and 1995 year-classes.

METHODS

Handling of collection — All otoliths and associated data were transferred to the Center for Quantitative Fisheries Ecology's Age and Growth Laboratory as they were collected. In the lab they were sorted by date of capture, their envelope labels verified against VMRC's collection data, and each fish was assigned a unique Age and Growth Laboratory sample number. All otoliths were stored dry in labeled cell well plates.

Preparation — Otoliths from fish were processed using an Age and Growth Laboratory thin section technique modified to deal with the fragile nature of Spanish mackerel otoliths. Briefly, an otolith was first embedded in a 9.5 mm x 4.5 mm x 4.5 mm silicon mold well with Loctite 349 photo-active adhesive. The mold was placed under ultraviolet light to cure and harden the Loctite. The embedded otolith was removed from the Silicon mold and the location of the core of the otolith was then marked with an extra fine point permanent marker. A thin transverse section was made using a Buelher Isomet saw equipped with two high concentration Norton diamond wafering blades separated by a 0.4 mm steel spacer. The otolith section was mounted best-side up onto a glass slide with Crystal Bond. The section was then lightly polished on a Buehler Ecomet 3 variable speed grinder-polisher with Mark V Laboratory 30-micron polishing film. The thin-section was then covered with a thin layer of Flo-texx mounting medium, which provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — By convention, a birth date of January 1 is assigned to all Northern Hemisphere fish species. We use a system of age determination that assigns age class according to the date of sacrifice with respect to this international accepted birth date and the timing of annulus formation. Although an otolith annulus is actually the combination of an opaque and translucent band, when ageing otoliths we actually enumerate only the opaque bands, but still refer to them as annuli. Spanish mackerel otolith annulus formation occurs between the months of April and June, with younger fish tending to lay down annuli earlier than older fish. Fish age is written first followed by the actual number of annuli visible listed within parentheses (e.g., 3(3)). The presence of a "+" after the number in the parentheses indicates new growth, or "plus growth" visible on the structure's margin. Using this method, a fish sacrificed in January before annulus formation with three visible annuli would be assigned the same age, 4(3+), as a fish with four visible annuli sacrificed in August after annulus formation, 4(4+). Year-class is then assigned once the reader determines the fish's age and takes into account the year of capture.

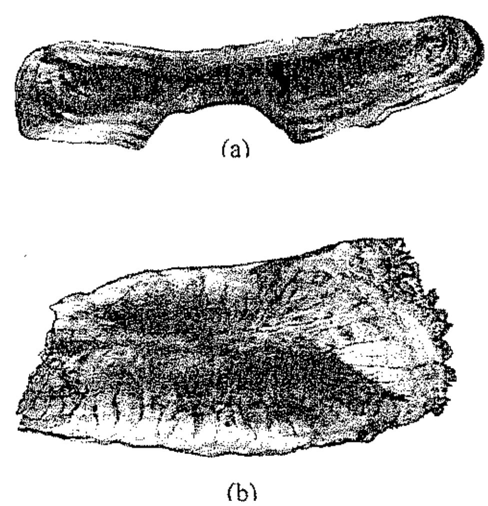


Figure 1. A three year old spanish mackerel otolith from a 0.6 kg male a) thin-section b) whole otolith with part of the tip broken off.

Two different readers aged all sectioned otoliths using a Leica MZ-12 dissecting microscope with polarized transmitted light at between 8 and 40 times magnification. The first annulus on sectioned otoliths was often quite distant from the core, with subsequent annuli regularly spaced along the sulcal groove out towards the proximal (inner-face) edge of the otolith (Figures 1 and 2).

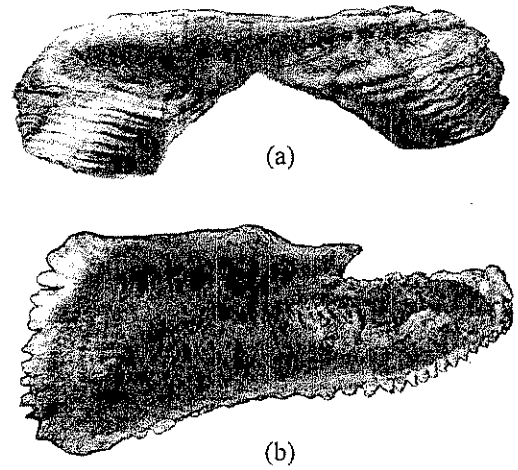


Figure 2. An eight year old spanish mackerel otolith from a 1 kg female a) thin-section b) whole otolith.

All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995).

RESULTS

The average between-reader coefficient of variation (CV) of 2.0% was considered not significant. Figure 3 illustrates the between readers' precision of age estimates. There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 6.29$, df $= 4$, $P = .17$).

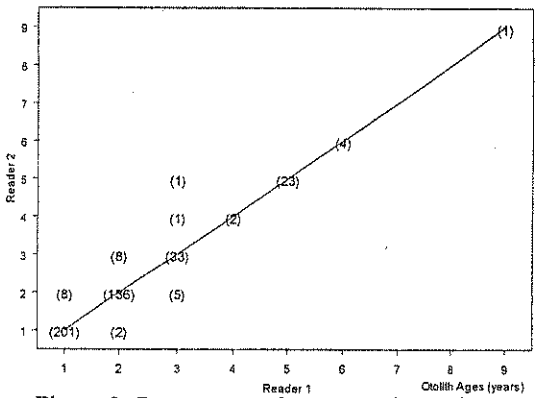


Figure 3. Between-reader comparison of otolith age estimates for Spanish mackerel.

Of the 425 Spanish mackerel aged with otoliths, seven age classes were represented (Table 3). The average age was 1.84 year old, and the standard deviation and standard error were 1.1 and 0.05, respectively. Year-class data (Figure 4) show that the fishery was comprised of seven year-classes, comprising fish from the 1995 and 1998 through 2003 year-classes, with fish primarily from the 2002 and 2003 year-classes.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

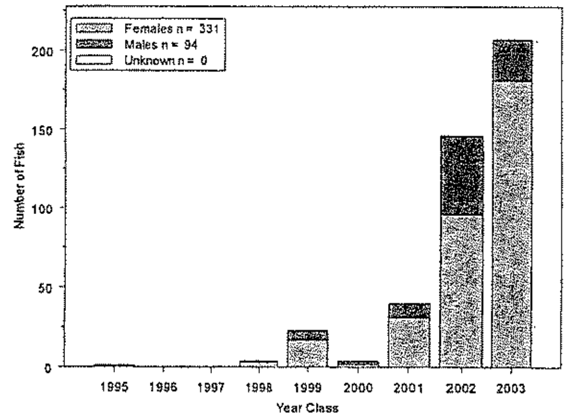


Figure 4. Year-class frequency distribution for Spanish mackerel collected for ageing in 2004. Distribution for otolith ages is broken down by sex.

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Kimura, D.K. 1980. Likelihood methods for the von Bertalanffy growth curve. Fish. Bull. 77:765-776.

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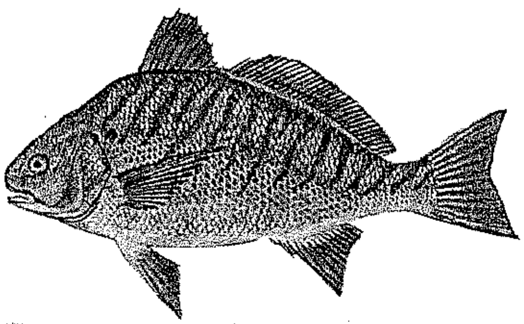
Table 1. The number of Spanish mackerel assigned to each total length-at-age category for 425 fish sampled for age determination in Virginia during 2004 (no length for 1 fish).

Length 1-inch intervals	Age (years)							
	1	2	3	4	5	6	0	Totals
13 - 13.99	2	1	0	0	0	0	0	3
14 - 14.99	15	0	0	0	0	0	0	15
15 - 15.99	83	2	0	0	0	0	0	85
16 - 16.99	71	11	0	0	0	0	0	82
17 - 17.99	25	26	6	0	0	0	0	57
18 - 18.99	9	25	5	0	0	0	0	39
19 - 19.99	2	38	10	0	1	1	0	52
20 - 20.99	0	24	7	1	1	1	0	34
21 - 21.99	0	12	2	3	2	0	0	19
22 - 22.99	0	6	3	0	4	0	0	13
23 - 23.99	0	1	2	0	3	0	0	6
24 - 24.99	0	0	1	0	4	0	0	5
25 - 25.99	0	0	3	0	5	0	0	8
26 - 26.99	0	0	0	0	3	0	0	3
27 - 27.99	0	0	0	0	0	1	0	1
28 - 28.99	0	0	0	0	1	1	0	2
Totals	207	146	39	4	24	4	0	424

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for Spanish mackerel sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)							
	1	2	3	4	5	6	0	N
13 - 13.99	0.667	0.333	0.000	0.000	0.000	0.000	0.000	3
14 - 14.99	1.000	0.000	0.000	0.000	0.000	0.000	0.000	15
15 - 15.99	0.976	0.024	0.000	0.000	0.000	0.000	0.000	85
16 - 16.99	0.866	0.134	0.000	0.000	0.000	0.000	0.000	82
17 - 17.99	0.439	0.456	0.105	0.000	0.000	0.000	0.000	57
18 - 18.99	0.231	0.641	0.128	0.000	0.000	0.000	0.000	39
19 - 19.99	0.038	0.731	0.192	0.000	0.019	0.019	0.000	52
20 - 20.99	0.000	0.706	0.206	0.029	0.029	0.029	0.000	34
21 - 21.99	0.000	0.632	0.105	0.158	0.105	0.000	0.000	19
22 - 22.99	0.000	0.462	0.231	0.000	0.308	0.000	0.000	13
23 - 23.99	0.000	0.167	0.333	0.000	0.500	0.000	0.000	6
24 - 24.99	0.000	0.000	0.200	0.000	0.800	0.000	0.000	5
25 - 25.99	0.000	0.000	0.375	0.000	0.625	0.000	0.000	8
26 - 26.99	0.000	0.000	0.000	0.000	1.000	0.000	0.000	3
27 - 27.99	0.000	0.000	0.000	0.000	0.000	1.000	0.000	1
28 - 28.99	0.000	0.000	0.000	0.000	0.500	0.500	0.000	2
						Total Sampled		424

Chapter 8
Spot



Leiostomus xanthurus

INTRODUCTION

A total of 459 spot, *Leiostomus xanthurus*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age for the sample was 2.0 year old, and the standard deviation and standard error were 0.69 and 0.03, respectively. Six age classes (1 to 6) were represented, comprising fish from the 1998-2003 year-classes, with fish predominantly from the 2002 year-class.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and assigned unique Age and Growth Laboratory sample numbers. All otoliths were stored dry in labeled cell well trays.

Preparation — Otoliths were processed for ageing using a thin-sectioning technique. The first step in otolith preparation was to grind down the otolith in a transverse plane to its core using a Hillquist thin section machine's 320-mesh diamond cup wheel. To prevent distortion of the reading surface, the otolith was ground exactly perpendicular to the reading plane. The ground side of the otolith was then placed face down in a drop of Loctite 349 photo-active adhesive on a labeled glass slide and placed under ultraviolet light to allow the adhesive to harden. The Hillquist thin section machine's cup wheel was used again to grind the otolith, embedded in Loctite, to a thickness of 0.3 to 0.5 mm. Finally, a thin layer of Flo-texx mounting medium was applied to the otolith section to increase light transmission through the translucent zones, which provided enhanced contrast and greater readability.

Readings — Two different readers aged all sectioned otoliths using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 1).

All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both

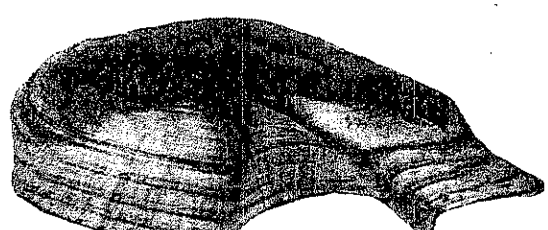


Figure 1. Sectioned otolith from a 5 year old spot.

readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. To detect any changes or drift in our ageing methods, both readers re-aged otoliths of 50 randomly selected fish previously aged in 2000. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

Measurements of reader precision were high, with age disagreements for only 17 out of 458 fish aged. Figure 2 illustrates the between readers' precision of age estimates. Both reader 1 and reader 2 had 100 % agreement for precision age reading. There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 7.34$, df = 3, $P = 0.06$). Figure 2 illustrates the between readers' precision of age estimates, with no differences greater than one year.

Of the 458 fish aged with otoliths, 6 age classes were represented (Table 1). The average age for the sample was 2.0 year old, and the standard deviation and standard error were 0.69 and 0.03, respectively.

Year-class data (Figure 3) show that the fishery was comprised of 6 year-classes, with fish spawned in 2002 dominating the catch.

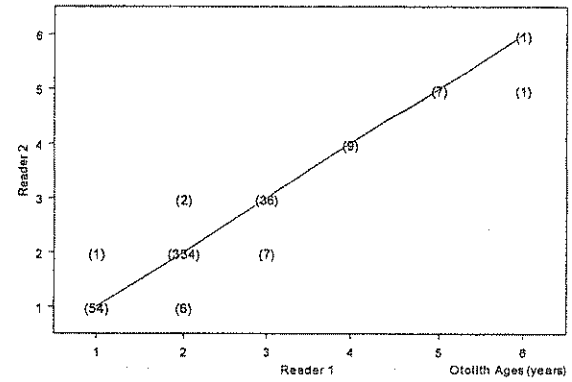


Figure 2. Between-reader comparison of otolith age estimates for spot.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

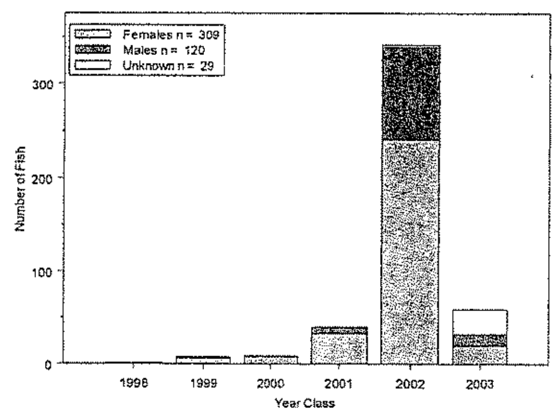


Figure 3. Year-class distribution for spot collected for ageing in 2004. Distribution is broken down by sex.

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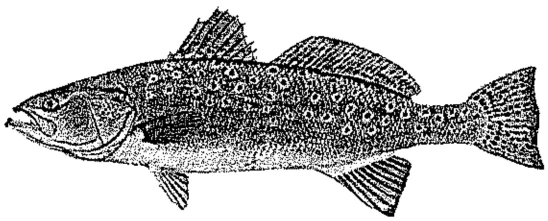
Table 1. The number of spot assigned to each total length-at-age category for 459 fish sampled for age determination in Virginia during 2004 (Length not reported for 3 fish).

Length	Age						
1-inch intervals	1	2	3	4	5	6	Totals
6 - 6.99	2	0	0	0	0	0	2
7 - 7.99	5	1	0	0	0	0	6
8 - 8.99	19	21	0	0	0	0	40
9 - 9.99	24	115	0	0	0	0	139
10 - 10.99	4	130	8	0	0	0	142
11 - 11.99	3	53	21	0	0	0	77
12 - 12.99	0	19	9	5	5	0	38
13 - 13.99	0	2	2	3	3	1	11
14 - 14.99	0	0	0	1	0	0	1
Totals	57	341	40	9	8	1	456

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for spot sampled for age determination in Virginia during 2004 (Length not reported for 3 fish).

Length	Age						
1-inch intervals	1	2	3	4	5	6	N
6 - 6.99	1.000	0.000	0.000	0.000	0.000	0.000	2
7 - 7.99	0.833	0.167	0.000	0.000	0.000	0.000	6
8 - 8.99	0.475	0.525	0.000	0.000	0.000	0.000	40
9 - 9.99	0.173	0.827	0.000	0.000	0.000	0.000	139
10 - 10.99	0.028	0.915	0.056	0.000	0.000	0.000	142
11 - 11.99	0.039	0.688	0.273	0.000	0.000	0.000	77
12 - 12.99	0.000	0.500	0.237	0.132	0.132	0.000	38
13 - 13.99	0.000	0.182	0.182	0.273	0.273	0.091	11
14 - 14.99	0.000	0.000	0.000	1.000	0.000	0.000	1
					Sample Size		456

Chapter 9
Spotted Seatrout



Cynoscion nebulosus

INTRODUCTION

A total of 501 spotted seatrout, *Cynoscion nebulosus*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age for the sample was 1.0 years old, and the standard deviation and standard error were 0.62 and 0.03, respectively. Four age classes (0 to 3) were represented, comprising fish from the 2001-2004 year-classes, with fish primarily from the 2003 year-class.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. They were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and each fish assigned a unique Age and Growth Laboratory sample number. All otoliths were stored dry in labeled cell well trays.

Preparation — The first step in seatrout otolith preparation was to make a transverse cut just off center of the otolith with a Hillquist thin section machine's cut-off saw equipped with an HCR-100 diamond blade. To prevent distortion of the reading surface, the cut surface of the otolith half containing the focus was ground down on a Hillquist thin section machine's 320 mesh diamond cup wheel until perpendicular to the reading plane. The otolith's ground surface was then placed face down in a drop of Loctite 349 photo-active adhesive on a labeled glass slide and placed under ultraviolet light to allow the adhesive to harden (approximately ten minutes). The Hillquist thin section machine's cup wheel was used again to grind the otolith, embedded in Loctite, to a thickness of 0.3 to 0.5 mm. Finally, a thin layer of Flo-texx mounting medium was applied to the otolith section to increase light transmission through the translucent zones, which provided enhanced contrast and greater readability.

Readings — Two different readers aged all sectioned otoliths using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 1). All

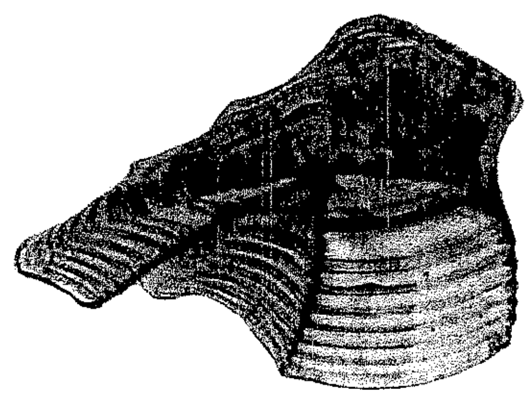


Figure 1. Sectioned otolith from an 8 year old male spotted seatrout.

samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, both readers aged all 50 fish a second time to measure reader precision and age reproducibility. To detect any changes or drift in our ageing methods, both readers re-aged otoliths of 50 randomly selected fish previously aged in 2000. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

No bias was discovered in any of the self-precision tests of otolith age estimates, with both readers equally able to reproduce the ages of previously read samples. There was also 100 percent agreement between reader age estimates. Figure 2 illustrates the between readers' precision of age estimates. There was no evidence of drift in age determination from the 2000 precision fish with 100% agreement for both readers.

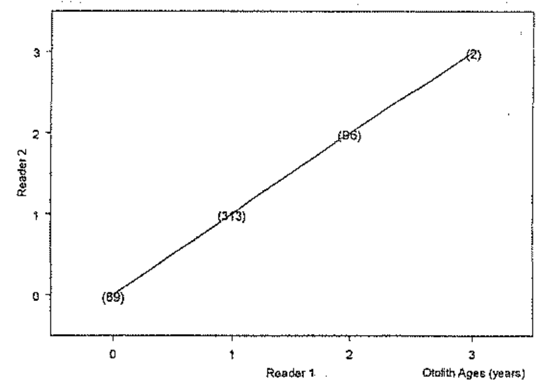


Figure 2. Between-reader comparison of otolith age estimates for spotted seatrout.

Of the 500 fish aged with otoliths, 4 age classes were represented (Table 1). The average age for the sample was 1.0 years old, and the standard deviation and standard error were 0.62 and 0.02, respectively.

Year-class data (Figure 3) show that the fishery was comprised of 4 year-classes, comprising fish from the 2001-2004 year-classes, with fish primarily from the 2003 year-class.

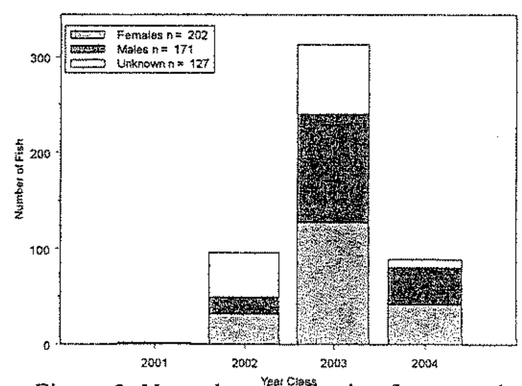


Figure 3. Year-class distribution for spotted seatrout collected for ageing in 2004. Distribution is broken down by sex.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

REFERENCES

Campana, S.E., M.C. Annand, and J.I. McMillan. 1995. Graphical and statistical methods for determining the consistency of age determinations. Trans. Am. Fish. Soc. 124:131-138.

Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

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Table 1. The number of spotted seatrout assigned to each total length-at-age category for 501 fish sampled for age determination in Virginia during 2004 (no length for 46 fish).

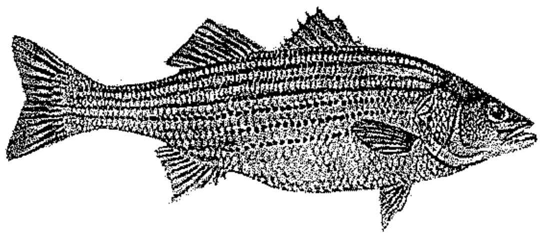
Length 1-inch intervals	Age (years)				
	0	1	2	3	Totals
4 - 4.99	1	0	0	0	1
5 - 5.99	1	0	0	0	1
6 - 6.99	1	0	0	0	1
7 - 7.99	1	0	0	0	1
8 - 8.99	2	0	0	0	2
9 - 9.99	5	0	0	0	5
10 - 10.99	29	1	0	0	30
11 - 11.99	29	11	0	0	40
12 - 12.99	19	18	0	0	37
13 - 13.99	0	37	0	0	37
14 - 14.99	0	51	1	0	52
15 - 15.99	0	50	2	0	52
16 - 16.99	0	35	7	0	42
17 - 17.99	0	31	13	0	44
18 - 18.99	0	25	11	0	36
19 - 19.99	0	14	8	0	22
20 - 20.99	0	2	23	0	25
21 - 21.99	0	0	9	1	10
22 - 22.99	0	0	12	0	12
23 - 23.99	0	0	1	0	1
24 - 24.99	0	0	1	0	1
25 - 25.99	0	0	1	0	1
26 - 26.99	0	0	0	1	1
Totals	88	275	89	2	454

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for spotted seatrout sampled for age determination in Virginia during 2004 (no length for 46 fish).

Length 1-inch intervals	Age (years) 0	1	2	3	N
4 - 4.99	1.000	0.000	0.000	0.000	1
5 - 5.99	1.000	0.000	0.000	0.000	1
6 - 6.99	1.000	0.000	0.000	0.000	1
7 - 7.99	1.000	0.000	0.000	0.000	1
8 - 8.99	1.000	0.000	0.000	0.000	2
9 - 9.99	1.000	0.000	0.000	0.000	5
10 - 10.99	0.967	0.033	0.000	0.000	30
11 - 11.99	0.725	0.275	0.000	0.000	40
12 - 12.99	0.514	0.486	0.000	0.000	37
13 - 13.99	0.000	1.000	0.000	0.000	37
14 - 14.99	0.000	0.981	0.019	0.000	52
15 - 15.99	0.000	0.962	0.038	0.000	52
16 - 16.99	0.000	0.833	0.167	0.000	42
17 - 17.99	0.000	0.705	0.295	0.000	44
18 - 18.99	0.000	0.694	0.306	0.000	36
19 - 19.99	0.000	0.636	0.364	0.000	22
20 - 20.99	0.000	0.080	0.920	0.000	25
22 - 22.99	0.000	0.000	1.000	0.000	12
23 - 23.99	0.000	0.000	1.000	0.000	1
24 - 24.99	0.000	0.000	1.000	0.000	1
25 - 25.99	0.000	0.000	1.000	0.000	1
26 - 26.99	0.000	0.000	0.000	1.000	1
			Sample Size		454

Chapter 10

Striped Bass



Morone saxatilis

INTRODUCTION

A total of 1396 striped bass, *Morone saxatilis*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2005. Only otoliths were collected from 1 fish and only scales were collected from 1068 fish, leaving 327 fish from which both scales and otoliths were collected with one specimen. The average scale age was 9.5 years, with 16 age classes (3 to 18) comprising fish from 1987 to 2002 year-classes. The average otolith age was 8.0 years, with 17 age classes (3 to 18 and 23) comprising fish from 1982 to 2002 year-classes.

METHODS

Handling of collection — Otoliths and scales were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and each fish assigned a unique Age and Growth Laboratory sample number. All otoliths were stored dry in labeled cell well plates, while scales were stored in their original coin envelopes.

Preparation —

Scales – Striped bass scales were prepared for age and growth analysis by making acetate impressions of the scale microstructure. Due to extreme variation in the size and shape of scales from individual fish, we selected only those scales that had even margins and uniform size. We selected a range of four to six preferred scales (based on overall scale size) from each fish, making sure that only non-regenerated scales were used. Scale impressions were made on extruded clear 020 acetate sheets (25 mm x 75 mm) with a Carver Laboratory Heated Press (model "C"). The scales were pressed with the following settings:

Pressure:	15000 psi
Temperature:	77°C (170°F)
Time:	5 to 10 min

Striped bass scales that were the size of a quarter (coin) or larger, were pressed individually for up to twenty minutes. After pressing, the impressions were viewed with a Bell and Howell microfiche reader and checked again for regeneration and incomplete margins. Impressions that were too light, or when all scales were regenerated a new impression was made using different scales from the same fish.

Otoliths — We used a thin-section and bake technique to process striped bass otoliths for age determination. Otolith preparation began by randomly selecting either the right or left otolith. The otolith was mounted with Crystal Bond onto a standard microscope slide with its distal surface orientated upwards. Once mounted, a small mark was placed on the otolith surface directly above the otolith focus. The slide, with attached otolith, was then secured to an Isomet saw equipped with two diamond wafering blades separated by a 0.5 mm spacer, which was slightly smaller in diameter than the diamond blades. The otolith was positioned so that the wafering blades straddled each side of the otolith focus ink mark. It was crucial that this cut be perpendicular to the long axis of the otolith. Failure to do so resulted in "broadening" and distortion of winter growth zones. A proper cut resulted in annuli that were clearly defined and delineated. Once cut, the otolith section was placed into a ceramic "Coors" spot plate well and baked in a Thermolyne 1400 furnace at 400°C. Baking time was otolith size dependent and gauged by color, with a light caramel color desired. Once a suitable color was reached the baked thin-section was placed on a labeled glass slide and covered with a thin layer of Flo-texx mounting medium, which provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — By convention, a birthdate of January 1 is assigned to all Northern Hemisphere fish species. We use a system of age determination that assigns age class according to the date of sacrifice with respect to this international accepted birthdate and the timing of annulus formation, which occurs between the months of May and June for striped bass. Once the reader decides how many annuli are visible on the ageing structure, the year class is assigned. The year class designation, or age, is written first followed by the actual number of annuli visible listed within brackets (e.g. 3(3)). The presence of a "+" after the number in the brackets indicates new growth, or "plus growth" visible on the structure's margin. Using this method, a fish sacrificed in January before annulus formation with three visible annuli would be assigned the same age, 4(3+), as a fish with four visible annuli sacrificed in July after annulus formation, 4(4).

Two different readers aged all samples in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the age readers were unable to agree on a final age, the fish was excluded from further analysis.

Scales - We determined fish age by viewing acetate impressions of scales (Figure 1) with a standard Bell and Howell R-735 microfiche reader equipped with 20 and 29 mm lenses.

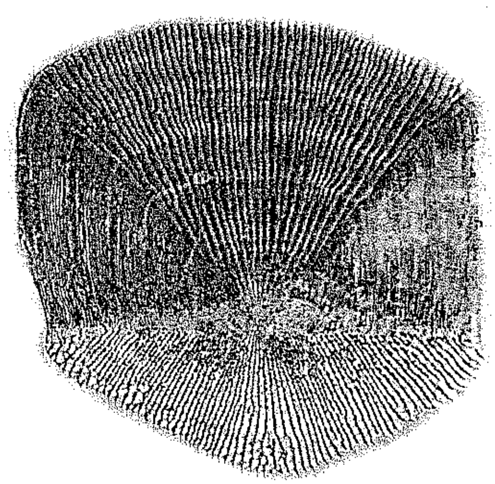


Figure 1. Scale impression of a 5-year-old male striped bass.

Annuli on striped bass scales are identified based on two scale microstructure features, "crossing over" and circuli disruption. Primarily, "crossing over" in the lateral margins near the posterior\anterior interface of the scale is used to determine the origin of the annulus. Here compressed circuli (annulus) "cross over" the previously deposited circuli of the previous year's growth. Typically annuli of the first three years can be observed transversing this interface as dark bands. These bands remain consistent throughout the posterior field and rejoin the posterior\anterior interface on the opposite side of the focus. Annuli can also be observed in the anterior lateral field of the scale. Here the annuli typically reveal a pattern of discontinuous and suddenly breaking segmented circuli. This event can also be distinguished by the presence of concentric white lines, which are typically associated with the disruption of circuli.

Annuli can also be observed bisecting the perpendicular plain of the radial striations in the anterior field of the scale. Radii emanate out from the focus of the scale towards the outer corner margins of the anterior field. These radial striations consist mainly of segmented concave circuli. The point of intersection between radii and annuli results in a "straightening out" of the concave circuli. This straightening of the circuli should be consistent throughout the entire anterior field of the scale. This event is further amplified by the presence of concave circuli neighboring both directly above and below the annulus.

The first year's annulus can be difficult to locate on some scales. It is typically best identified in the lateral field of the anterior portion of the scale. The distance from the focus to the first year's annulus is typically larger with respect to the following few annuli. For the annuli two through six, summer growth generally decreases proportionally. For ages greater than six, a crowding effect of the annuli near the outer margins of the scale is observed. This crowding effect creates difficulties in edge interpretation. At this point it is best to focus on the straightening of the circuli at the anterior margins of the scale.

When ageing young striped bass, zero through age two, extreme caution must be taken as not to over age the structure. Young fish have no point of reference to aid in the determination of the first year; this invariably results in over examination of the scale and such events as hatching or saltwater incursion marks (checks) may be interpreted as the first year.

Otoliths – Sectioned otoliths were aged by two different readers using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 2).

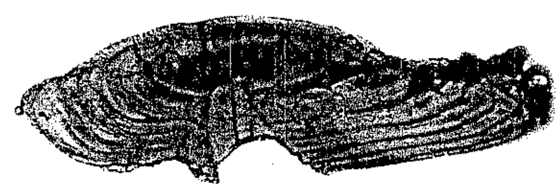


Figure 2. Otolith thin-section of a 5-year-old male striped bass.

By convention an annulus is identified as the narrow opaque zone, or winter growth. Typically the first year's annulus can be determined by first locating the focus of the otolith. The focus is generally located, depending on preparation, in the center of the otolith, and is visually well defined as a dark oblong region. The first year's annulus can be located directly below the focus, along the outer ridge of the sulcal groove on the ventral and dorsal sides of the otolith. This insertion point along the sulcal ridge resembles a check mark (not to be confused with a false annulus). Here the annulus can be followed outwards along the ventral and dorsal surfaces where it encircles the focus.

Subsequent annuli also emanate from the sulcal ridge, however, they do not encircle the focus, but rather travel outwards to the distal surface of the otolith. To be considered a true annulus, each annulus must be rooted in the sulcus and travel without interruption to the distal surface of the otolith. The annuli in striped bass have a tendency to split as they advance towards the distal surface. As a result, it is critical that reading path proceed in a direction down the sulcal ridge and outwards to the distal surface.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

Scales — Measurements of reader self-precision was marginal; with both readers able to reproduce the ages of previously read scales (reader 1's CV = 9.6% and reader 2's CV = 5.3%). In Figure 3 we present a graph of the results for between-reader scale ageing precision. There was good between-reader agreement for scale age readings, with age differences between the two readers one year or less for 78.1% of all aged fish. The average between-reader coefficient of variation (CV) of 7.4% was not significant, and comparable to the CV of 5.4% from 2004. There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 48.8$, df = 37, $P = 0.09$).

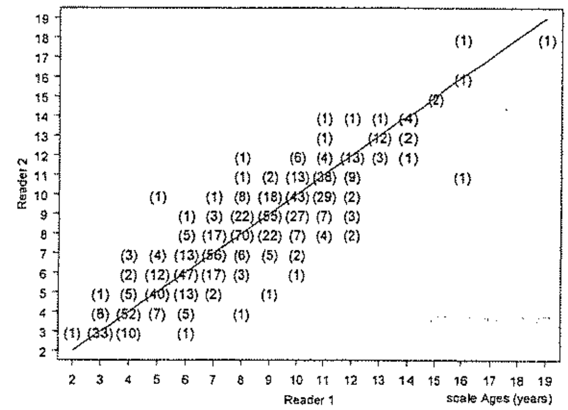


Figure 3. Between-reader comparison of scale age estimates for striped bass.

Of the 1395 striped bass aged with scales, 16 age classes (3 to 18) were represented. The average age for the sample was 9.4 years. The standard deviation and standard error were 2.8 and 0.10, respectively.

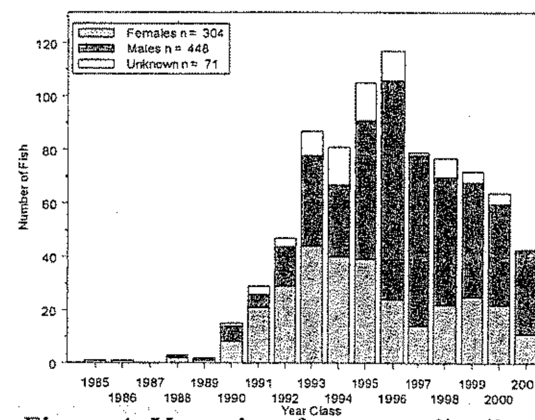


Figure 4. Year-class frequency distribution for striped bass collected for ageing in 2004. Distribution of scale ages is broken down by sex.

Year-class data (Figure 4) indicates that recruitment into the fishery typically begins at age 3, which corresponds to the 2001 year-class for striped bass caught in 2004. Striped bass appear to fully recruit to the fishery at age 8 (1996 year-class).

Otoliths — There was good between-reader agreement for otolith age readings using sectioned otoliths, with age differences between the two readers one year or less for 98.0% of all aged fish (Figure 5). The between reader average CV for otolith age estimates was only 1.69%, very comparable to the CV of 1.52% reported for 2003 fish. Like scale ages, there was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 14.23$, df = 19, $P = 0.7$).

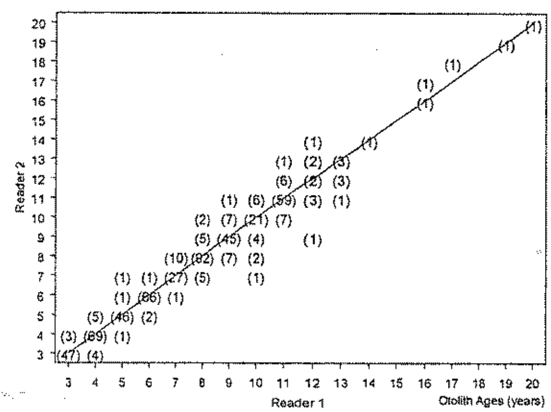


Figure 5. Between-reader comparison of otolith age estimates for striped bass.

Measurements of reader self-precision were high, with both readers able to reproduce the ages of previously read otoliths (Reader 1's CV = 4.1% and Reader 2's CV = 0.9%). Eighteen age classes (3 to 20) were represented for striped bass aged with otoliths. The average age for the sample was 7.3 years. The standard deviation and standard error were 2.8 and 0.11, respectively.

Comparison of Scale and Otolith Ages — While the CV of otolith and scales age estimates was 9.0%, there was also significant evidence of systematic disagreement between otolith and scale ages (test of symmetry, $\chi^2 = 94.2$, df = 45, $P <$ 0.01). Scales were assigned a lower age than otoliths for 26% of the fish and 32% of the time were scales assigned a higher age than otoliths (Figure 6). There was also evidence of bias between otolith and scale ages using an age bias plot (Figure 7), again with scales generally assigned higher ages for younger fish and lower ages for older fish than otoliths age estimates.

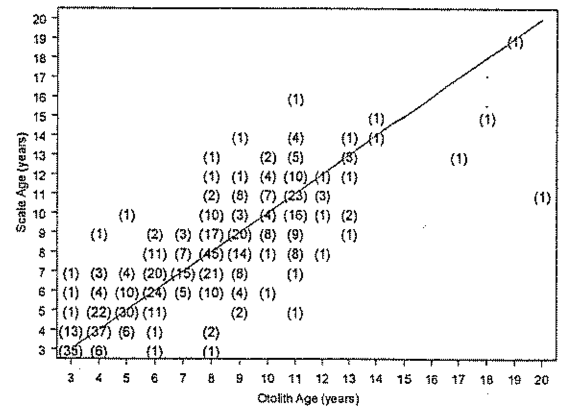


Figure 6. Comparison of otolith and scale age estimates for striped bass.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using scale ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

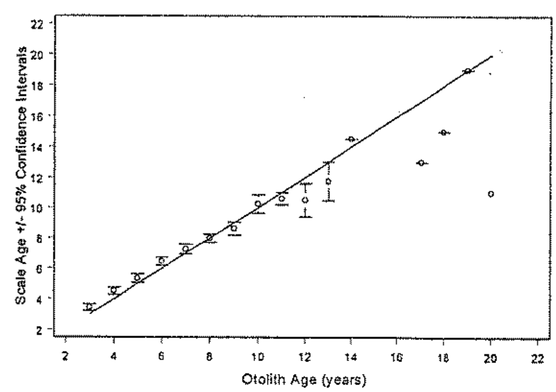


Figure 7. Age-bias plot for striped bass scale and otolith age estimates.

RECOMMENDATIONS

• We recommend that VMRC and ASMFC use otoliths for ageing striped bass. Although there is more preparation time for otoliths compared to scales, as the mean age of striped bass increases in the recovering fishery, otoliths should provide more reliable estimates of age. We will continue to compare the age estimates between otoliths and scales.

REFERENCES

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Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

S-PLUS. 1999. Guide to Statistics, Vol 1. Data Analysis and Products Division. MathSoft, Inc. Seattle, Washington.

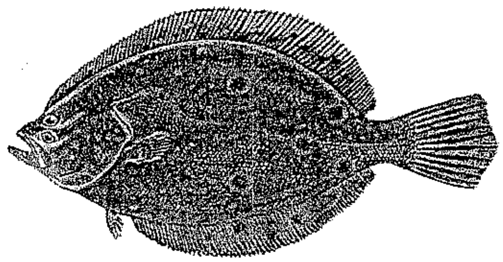
Table 1. The number of striped bass assigned to each total length-at-age category for fish collected for age determination in Virginia during 2004 (length not determined for 12 fish).

Length 1-inch intervals	Age (years)																
	3	4	5	6	7	8	9	10	11	12	13	14	15	16	18	19	Totals
17 - 17.99	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3
18 - 18.99	9	5	1	0	0	0	0	0	0	0	0	0	0	0	0	0	15
19 - 19.99	14	9	2	1	0	0	0	0	0	0	0	0	0	0	0	0	26
20 - 20.99	8	8	7	4	1	0	1	0	0	0	0	0	0	0	0	0	29
21 - 21.99	3	20	12	7	8	3	2	0	0	0	0	0	0	0	0	0	55
22 - 22.99	2	10	16	16	12	18	1	1	1	0	0	0	0	0	0	0	77
23 - 23.99	4	6	12	16	13	22	7	2	2	0	0	0	0	0	0	0	84
24 - 24.99	0	5	9	11	14	15	6	3	3	0	0	0	0	0	0	0	66
25 - 25.99	0	1	4	9	11	10	7	4	2	0	0	0	0	0	0	0	48
26 - 26.99	0	0	4	4	6	10	6	7	1	2	0	0	0	0	0	0	40
27 - 27.99	0	0	3	2	5	5	7	2	6	2	1	2	0	0	0	0	35
28 - 28.99	0	0	1	3	1	3	3	6	6	0	0	0	0	0	0	0	23
29 - 29.99	0	0	0	3	1	4	6	1	4	2	0	2	0	1	0	0	24
30 - 30.99	0	0	0	0	1	5	14	2	3	3	0	0	0	0	0	0	28
31 - 31.99	0	0	0	1	2	7	10	4	3	0	0	2	0	1	0	0	30
32 - 32.99	0	0	0	0	2	5	6	6	7	1	1	0	0	0	0	0	28
33 - 33.99	0	0	0	0	1	3	13	13	2	3	2	0	0	0	0	0	37
34 - 34.99	0	0	0	0	0	3	5	5	11	5	3	0	0	0	0	0	32
35 - 35.99	0	0	0	0	0	3	5	9	13	10	4	2	0	0	0	0	46
36 - 36.99	0	0	0	0	0	1	4	10	8	5	7	2	0	0	0	0	37
37 - 37.99	0	0	0	0	0	0	2	4	7	9	3	0	0	1	0	0	26
38 - 38.99	0	0	0	0	0	0	0	2	3	1	4	2	1	0	0	0	13
39 - 39.99	0	0	0	0	0	0	0	0	2	2	1	0	0	0	0	0	5
40 - 40.99	0	0	0	0	0	0	0	0	1	2	0	0	0	0	0	0	3
41 - 41.99	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	1
42 - 42.99	0	0	0	0	0	0	0	0	1	0	0	1	1	0	1	0	4
43 - 43.99	0	0	0	0	0	0	0	0	0	0	1	1	0	0	0	0	2
44 - 44.99	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	1
45 - 45.99	0	0	0	0	0	0	0	0	1	0	1	0	0	0	0	0	2
48 - 48.99	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1
Totals	40	64	71	77	78	117	105	81	87	47	29	15	2	3	1	1	818

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on scale ages for striped bass sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years) 3	4	5	6	7	8	9	10	11	12	13	14	15	16	18	19	N
17 - 17.99	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	3
18 - 18.99	0.600	0.333	0.067	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	15
19 - 19.99	0.538	0.346	0.077	0.038	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	26
20 - 20.99	0.276	0.276	0.241	0.138	0.034	0.000	0.034	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	29
21 - 21.99	0.055	0.364	0.218	0.127	0.145	0.055	0.036	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	55
22 - 22.99	0.026	0.130	0.208	0.208	0.156	0.234	0.013	0.013	0.013	0.000	0.000	0.000	0.000	0.000	0.000	0.000	77
23 - 23.99	0.048	0.071	0.143	0.190	0.155	0.262	0.083	0.024	0.024	0.000	0.000	0.000	0.000	0.000	0.000	0.000	84
24 - 24.99	0.000	0.076	0.136	0.167	0.212	0.227	0.091	0.045	0.045	0.000	0.000	0.000	0.000	0.000	0.000	0.000	66
25 - 25.99	0.000	0.021	0.083	0.188	0.229	0.208	0.146	0.083	0.042	0.000	0.000	0.000	0.000	0.000	0.000	0.000	48
26 - 26.99	0.000	0.000	0.100	0.100	0.150	0.250	0.150	0.175	0.025	0.050	0.000	0.000	0.000	0.000	0.000	0.000	40
27 - 27.99	0.000	0.000	0.086	0.057	0.143	0.143	0.200	0.057	0.171	0.057	0.029	0.057	0.000	0.000	0.000	0.000	35
28 - 28.99	0.000	0.000	0.043	0.130	0.043	0.130	0.130	0.261	0.261	0.000	0.000	0.000	0.000	0.000	0.000	0.000	23
29 - 29.99	0.000	0.000	0.000	0.125	0.042	0.167	0.250	0.042	0.167	0.083	0.000	0.083	0.000	0.042	0.000	0.000	24
30 - 30.99	0.000	0.000	0.000	0.000	0.036	0.179	0.500	0.071	0.107	0.107	0.000	0.000	0.000	0.000	0.000	0.000	28
31 - 31.99	0.000	0.000	0.000	0.033	0.067	0.233	0.333	0.133	0.100	0.000	0.000	0.067	0.000	0.033	0.000	0.000	30
32 - 32.99	0.000	0.000	0.000	0.000	0.071	0.179	0.214	0.214	0.250	0.036	0.036	0.000	0.000	0.000	0.000	0.000	28
33 - 33.99	0.000	0.000	0.000	0.000	0.027	0.081	0.351	0.351	0.054	0.081	0.054	0.000	0.000	0.000	0.000	0.000	37
34 - 34.99	0.000	0.000	0.000	0.000	0.000	0.094	0.156	0.156	0.344	0.156	0.094	0.000	0.000	0.000	0.000	0.000	32
35 - 35.99	0.000	0.000	0.000	0.000	0.000	0.065	0.109	0.196	0.283	0.217	0.087	0.043	0.000	0.000	0.000	0.000	46
36 - 36.99	0.000	0.000	0.000	0.000	0.000	0.027	0.108	0.270	0.216	0.135	0.189	0.054	0.000	0.000	0.000	0.000	37
37 - 37.99	0.000	0.000	0.000	0.000	0.000	0.000	0.077	0.154	0.269	0.346	0.115	0.000	0.000	0.038	0.000	0.000	26
38 - 38.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.154	0.231	0.077	0.308	0.154	0.077	0.000	0.000	0.000	13
39 - 39.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.400	0.400	0.200	0.000	0.000	0.000	0.000	0.000	5
40 - 40.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.333	0.667	0.000	0.000	0.000	0.000	0.000	0.000	3
41 - 41.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	1
42 - 42.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.250	0.000	0.000	0.250	0.250	0.000	0.250	0.000	4
43 - 43.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.500	0.500	0.000	0.000	0.000	0.000	2
44 - 44.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	0.000	0.000	0.000	1
45 - 45.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.500	0.000	0.500	0.000	0.000	0.000	0.000	0.000	2
48 - 48.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	1
														Sample size			818

Chapter 11
Summer Flounder



Paralichthys dentatus

INTRODUCTION

A total of 380 summer flounder, *Paralichthys dentatus*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. Only otoliths were collected from 9 fish and only scales were collected from 4 fish, leaving 367 fish for which both scales and otoliths were collected. The average scale age was 2.8 years, representing 11 year-classes (1994 to 2004). Fish from the 2002-2003 year-classes dominated the collection. The average otolith age was 2.7 years, representing 11 year-classes (1994 to 2004).

METHODS

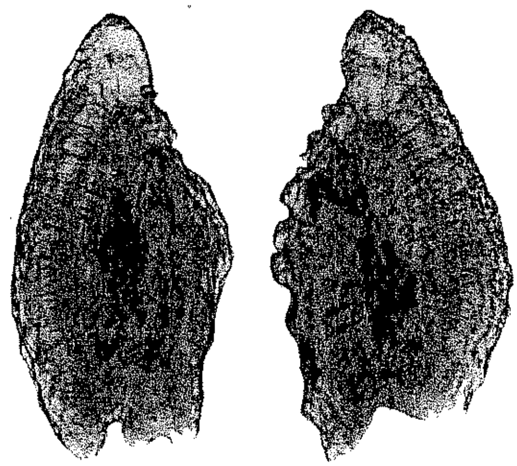
Handling of collection — Otoliths and scales were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and each fish assigned a unique Age and Growth Laboratory sample number. All otoliths were stored dry in labeled cell well plates, while scales were stored in their original coin envelopes.

Preparation —

Scales – Summer flounder scales were prepared for age and growth analysis by making acetate impressions of the scale microstructure. Due to extreme variation in the size and shape of scales from individual fish, we selected only those scales that had even margins and uniform size. We selected a range of five to ten preferred scales (based on overall scale size) from each fish, making sure that only non-regenerated scales were used. Scale impressions were made on extruded clear 020 acetate sheets (25 mm x 75 mm) with a Carver Laboratory Heated Press (model "C"). The scales were pressed with the following settings:

Pressure:	12000 to 15000 psi
Temperature:	Room temperature
Time:	7 minutes

Otoliths – The left otoliths of summer flounder are symmetrical in relation to the otolith nucleus, while right otoliths are asymmetrical (Figure 1). The right sagittal otolith was mounted with Aremco's clear Crystal Bond™ 509 adhesive onto a standard microscope slide with its distal surface orientated upwards. Once mounted, a small mark was placed on the otolith surface directly above the otolith focus. The slide, with attached otolith, was then secured to a Buehler Isomet saw equipped with two Norton diamond wafering blades separated by a 0.5 mm stainless steel spacer, which was slightly smaller in diameter than the diamond blades. The otolith was positioned so that the wafering blades straddled each side of the otolith focus ink mark. It was crucial that this cut be perpendicular to the long axis of the otolith.



(a) (b)

Figure 1. Whole otoliths from a 485 mm (total length) female summer flounder. (a) left otolith (b) right otolith.

Failure to do so resulted in "broadening" and distortion of winter growth zones. A proper cut resulted in annuli that were clearly defined and delineated. Once cut, the otolith section was placed into a ceramic "Coors" spot plate well and baked in a Thermolyne 1400 furnace at 400°C. Baking time was otolith size dependent and gauged by color, with a light caramel color desired. Once a suitable color was reached the baked thin-section was placed on a labeled glass slide and covered with a thin layer of Flo-texx mounting medium, which provided enhanced contrast and greater readability by increasing light transmission through the sections.

Readings — By convention, a birthdate of January 1 is assigned to all Northern Hemisphere fish species. The Age and Growth Lab uses a system of age determination that assigns age class according to the date of sacrifice with respect to this international accepted birthdate and the timing of annulus formation, which occurs in Virginia's waters between the months of February and April. Using this method, a fish sacrificed in January before annulus formation with three visible annuli will be assigned the same age as a fish with four visible annuli sacrificed in July after annulus formation. Once the reader has decided how many annuli are visible on the ageing structure, the year class is assigned. The year class designation, or age, is written first followed by the actual number of annuli visible listed within brackets (e.g. 3(3)). The presence of a "+" after the number in the brackets indicates new growth, or "plus growth" visible on the structure's margin.

Two different readers aged all samples in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Scales - We determined fish age by viewing the acetate impressions of scales (Figure 2) with a standard Bell and Howell R-735 microfiche reader equipped with 20 and 29 mm lenses.

Annuli on summer flounder scales are primarily identified by the presence of crossing over of circuli. Crossing over is most evident on the lateral margins near the posterior/anterior interface of the scale. Here compressed circuli (annulus) "cross over" the deposited circuli of the previous year's growth. Typically the annulus will protrude partially into the ctenii of the posterior field, but not always.

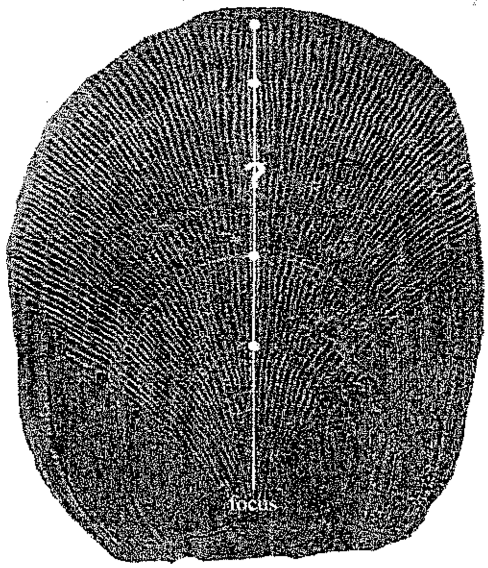


Figure 2. Scale impression of a 590 mm female summer flounder collected in November and aged as 4-years-old with scales. The question mark is located at a possible "3rd" annulus.

Following the annulus up into the anterior field of the scale reveals a pattern of discontinuous and suddenly breaking segmented circuli. This event can also be distinguished by the presence of concentric white lines, which are associated with the disruption of circuli. This pattern should be continuous throughout the entire anterior field of the scale. Locating the first annulus can be difficult due to latitudinal differences in growth rates and changes in the size of the first annulus due to a protracted spawning season. We consider the first annulus to be the first continuous crossing over event formed on the scale.

Otoliths – Sectioned otoliths were aged by two different readers using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 3).

Summer flounder otoliths are composed of visually distinct summer and winter growth zones. By convention, an annulus is identified as the narrow opaque zone, or winter growth band. With sectioned otoliths, to be considered a true annulus, these growth bands must be rooted in the sulcus and able to be followed, without interruption to the distal surface of the otolith. The annuli in summer flounder have a tendency to split as they advance towards the distal surface. As a result, it is critical that the reading path proceeds in a direction from the sulcus to the proximal surface. The first annulus is located directly below the focus and near the upper portion of the sulcal groove. The distance from the focus to the first year is moderate, with translucent zone deposition gradually becoming smaller as consecutive annuli are deposited towards the outer edge.

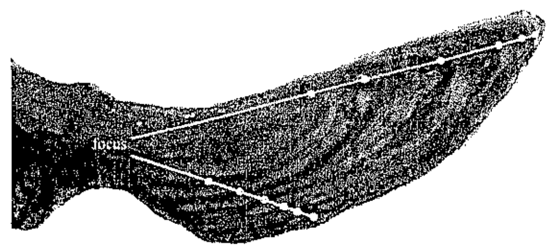


Figure 3. Otolith section from a 590 mm, 6-year-old female summer flounder collected in November. Same fish as Figure 2.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995).. A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

Scales — Measurements of reader self-precision were high, with both readers able to reproduce the ages of previously read scales (reader 1's CV = 5.3% and reader 2's CV = 1.6%). There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 13.1$, df = 8, P = 0.1). In Figure 4 we present a graph of the results for between-reader scale ageing precision. The average between-reader coefficient of variation (CV) of 3.3% was not significant.

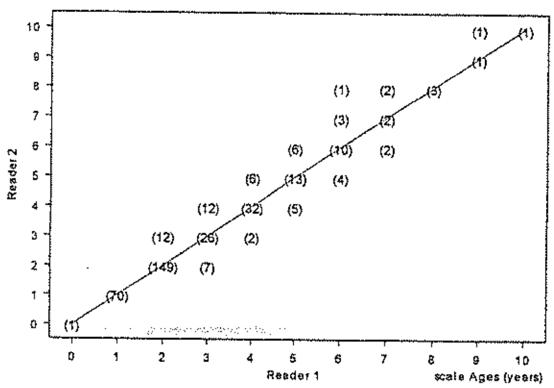


Figure 4. Between-reader comparison of scale age estimates for summer flounder.

Of the 371 fish aged with scales, 11 age-classes (0 to 10) were represented (Table 1). The average scale age was 2.7 years, and the standard deviation and standard error were 1.7 and 0.09, respectively.

Year-class data (Figure 5) indicate that recruitment into the fishery began at age 1, which corresponds to the 2003 year-class for summer flounder caught in 2004. Year-class abundance was high for the 2002–2003 year-classes, but declined sharply in the 2001 year-class and remained low for the earlier years.

Otoliths — Measurements of reader self-precision were high, with both readers able to reproduce the ages of previously read otoliths (reader 1's CV = 3.42% and reader 2's CV = 0.62%). There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 17$, df = 10, P = 0.07). In Figure 6 we present a graph of the results for between-reader otolith ageing precision. The average between-reader coefficient of variation (CV) of 1.12% was not significant.

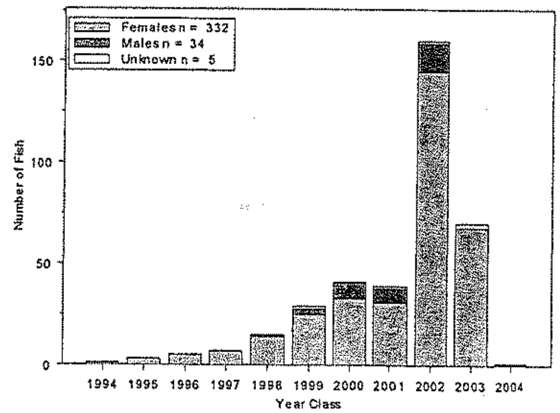


Figure 5. Scale year-class distribution for summer flounder collected in 2004. Distribution is broken down by sex.

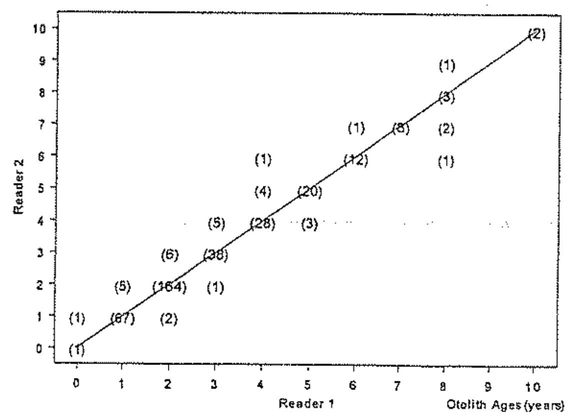


Figure 6. Between-reader comparison of otolith age estimates for summer flounder.

Of the 376 fish aged with otoliths, 11 age-classes (0 to 10) were represented. The average age for the sample was 2.7 years.

The standard deviation and standard error were 1.7 and 0.09, respectively.

Comparison of Scale and Otolith Ages — Otolith and scales ages were similar, with an average CV of 5.1% for the 367 fish for which both otoliths and scales were collected. Although statistically there was no evidence of systematic disagreement between otolith and scale ages (test of symmetry, $\chi^2 = 15.5$, df $= 15$, $P = 0.41$), signs of under-aging occurred and could be important when older year classes might be present. In Figure 7 we present a graph of the results for between-reader otolith/scale ageing precision. There was some evidence of bias between otolith and scale ages for the oldest fish in the sample (Figure 8), but this could be due to the extremely small number of fish in these age categories.

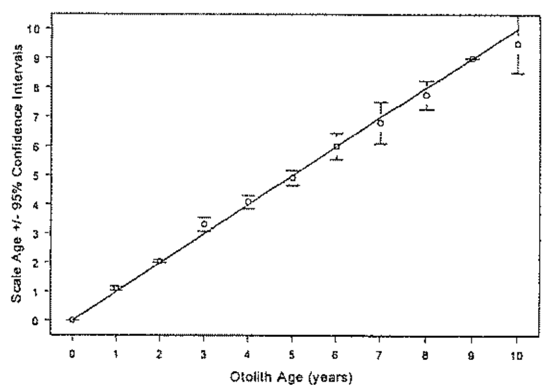


Figure 8. Age-bias plot for summer flounder scale and otolith age estimates.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using scale ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

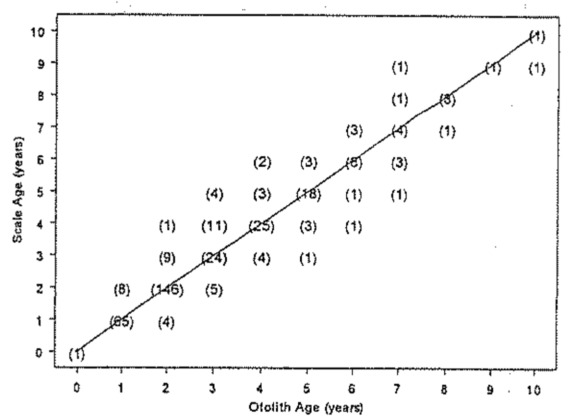


Figure 7. Comparison of otolith and scale age estimates for summer flounder.

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Hoenig, J.M., M.J. Morgan, and C.A. Brown. 1995. Analysing differences between two age determination methods by tests of symmetry. Can. J. Fish. Aquat. Sci. 52:364-368.

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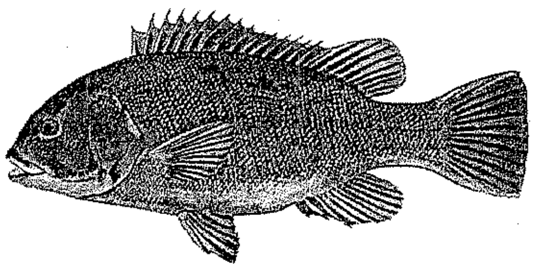
Table 1. The number of summer flounder assigned to each total length-at-age category for 380 fish sampled for age determination in Virginia during 2004 (scales not collected for 9 fish).

Length 1-inch intervals	Age (years)											
	0	1	2	3	4	5	6	7	8	9	10	Totals
11 - 11.99	0	2	0	0	0	0	0	0	0	0	0	2
12 - 12.99	0	11	1	0	0	0	0	0	0	0	0	12
13 - 13.99	0	14	4	1	0	0	0	0	0	0	0	19
14 - 14.99	1	28	33	4	2	0	0	0	0	0	0	68
15 - 15.99	0	13	58	5	2	0	0	1	0	0	0	79
16 - 16.99	0	2	35	6	7	3	0	0	0	0	0	53
17 - 17.99	0	0	18	7	9	3	1	0	0	0	0	38
18 - 18.99	0	0	6	7	6	3	0	0	0	0	0	22
19 - 19.99	0	0	2	4	10	3	0	0	0	0	0	19
20 - 20.99	0	0	1	2	2	6	1	0	0	0	0	12
21 - 21.99	0	0	2	0	1	6	5	0	0	0	0	14
22 - 22.99	0	0	0	1	1	2	1	0	1	0	0	6
23 - 23.99	0	0	0	2	1	1	1	3	0	0	0	8
24 - 24.99	0	0	0	0	0	1	5	3	0	1	0	10
25 - 25.99	0	0	0	0	0	0	2	1	1	0	0	4
26 - 26.99	0	0	0	0	0	0	0	0	1	1	1	3
27 - 27.99	0	0	0	0	0	0	0	0	1	0	0	1
29 - 29.99	0	0	0	0	0	0	0	0	0	1	0	1
Totals	1	70	160	39	41	28	16	8	4	3	1	371

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on scale ages for summer flounder sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years)											
	0	1	2	3	4	5	6	7	8	9	10	N
11 - 11.99	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	2
12 - 12.99	0.000	0.917	0.083	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	12
13 - 13.99	0.000	0.737	0.211	0.053	0.000	0.000	0.000	0.000	0.000	0.000	0.000	19
14 - 14.99	0.015	0.412	0.485	0.059	0.029	0.000	0.000	0.000	0.000	0.000	0.000	68
15 - 15.99	0.000	0.165	0.734	0.063	0.025	0.000	0.000	0.013	0.000	0.000	0.000	79
16 - 16.99	0.000	0.038	0.660	0.113	0.132	0.057	0.000	0.000	0.000	0.000	0.000	53
17 - 17.99	0.000	0.000	0.474	0.184	0.237	0.079	0.026	0.000	0.000	0.000	0.000	38
18 - 18.99	0.000	0.000	0.273	0.318	0.273	0.136	0.000	0.000	0.000	0.000	0.000	22
19 - 19.99	0.000	0.000	0.105	0.211	0.526	0.158	0.000	0.000	0.000	0.000	0.000	19
20 - 20.99	0.000	0.000	0.083	0.167	0.167	0.500	0.083	0.000	0.000	0.000	0.000	12
21 - 21.99	0.000	0.000	0.143	0.000	0.071	0.429	0.357	0.000	0.000	0.000	0.000	14
22 - 22.99	0.000	0.000	0.000	0.167	0.167	0.333	0.167	0.000	0.167	0.000	0.000	6
23 - 23.99	0.000	0.000	0.000	0.250	0.125	0.125	0.125	0.375	0.000	0.000	0.000	8
24 - 24.99	0.000	0.000	0.000	0.000	0.000	0.100	0.500	0.300	0.000	0.100	0.000	10
25 - 25.99	0.000	0.000	0.000	0.000	0.000	0.000	0.500	0.250	0.250	0.000	0.000	4
26 - 26.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.333	0.333	0.333	3
27 - 27.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	0.000	1
29 - 29.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	1
										Sample Size		371

Chapter 12
Tautog



Tautoga onitis

INTRODUCTION

A total of 506 tautog, *Tautoga onitis*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. Otoliths were not collected from 5 fish and opercula were not collected from 62 fish, leaving 439 fish for which both otoliths and opercula were collected. Our results and analyses are based on operculum ages, unless otherwise noted, to allow our data to be directly comparable to other tautog age and growth studies. The average operculum age for the sample was 4.7 years, and the standard deviation and standard error were 2.6 and 0.1, respectively. Fifteen age-classes (2-15, and 17) were represented, comprising fish from the 1987 and 1989 through 2002 year-classes.

METHODS

Handling of collection — Otoliths and opercula were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and each fish assigned a unique Age and Growth Laboratory sample number. All otoliths were stored dry in labeled cell well plates, while opercula were stored frozen in their original coin envelopes until processed.

Preparation —

Opercula – Tautog opercula were boiled for several minutes to remove any attached skin and muscle tissue. After boiling, opercula were examined to determine whether they were collected whole or in some way damaged. Opercula were allowed to dry and finally stored in new labeled coin envelopes.

Otoliths – Because of the small size of a tautog otolith, it required extra steps in preparation for ageing. An otolith was first baked in a Thermolyne 1400 furnace at 400°C for one to two minutes until it turned a medium brown color (caramel). The location of the core of the otolith was marked with a felt pen and the entire otolith was embedded in Loctite 349 adhesive, placed under UV light, and allowed to harden overnight. The otolith was then transversely sectioned through the felt pen mark with a low speed Buehler Isomet saw equipped with double wafering blades separated by a 0.5 mm spacer. The sectioned side of the otolith was then placed face down in a drop of Loctite 349 photo-active adhesive on a labeled glass slide and placed under ultraviolet light to allow the adhesive to harden (approximately ten minutes). The otolith section was then polished using a Buehler Ecomet 3 variable speed grinder-polisher with Mark V Laboratory 30-micron polishing film. After polishing, a thin layer of Flo-texx mounting medium was applied to the otolith section to increase light transmission through the translucent zones, which provided enhanced contrast and greater readability.

Readings — Opercula were aged on a light table with no magnification (Figure 1). Sectioned otoliths were aged by two different readers using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 2).

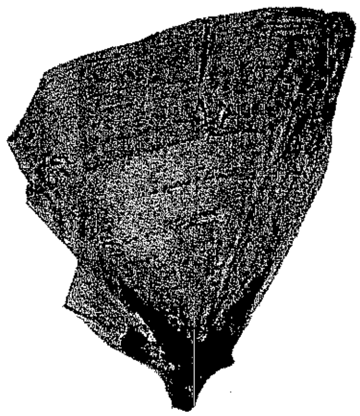


Figure 1. Operculum from a 13 year-old male tautog.

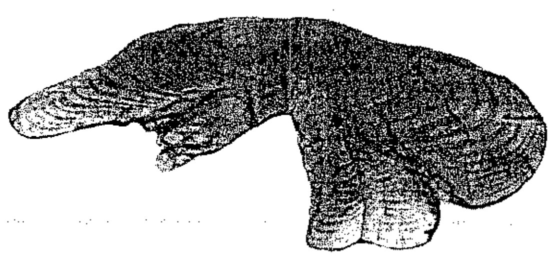


Figure 2. Otolith section from a 13 year-old male tautog. Same fish as Figure 1.

Two different readers aged all samples in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing

RESULTS

Opercula — Measurements of reader self-precision were high, with both readers able to reproduce the ages of previously read opercula (Reader 1's CV = 7.6% and Reader 2's CV = 2.6%). In Figure 3 we present a graph of the results for between-reader operculum ageing precision. There was evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 107$, df = 40, $P < 0.01$).

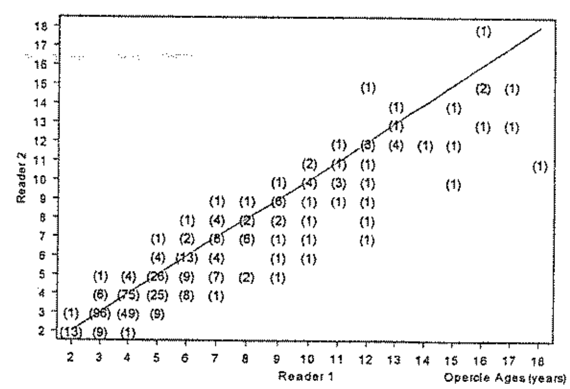


Figure 3. Between-reader comparison of operculum age estimates for tautog.

The average between-reader coefficient of variation (CV) of 8.3% and was not significant.

The average operculum age for the sample was 4.7 years, and the standard deviation and standard error were 2.6 and 0.1, respectively.

Year-class data (Figure 4) indicate that full recruitment into the fishery occurred at age three, which corresponds to the 2001 year-class for tautog caught in 2004. Year-class abundance was high for the 1999–2001 year-classes.

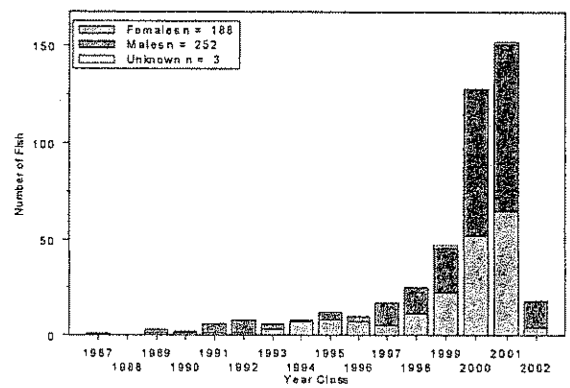


Figure 4. Operculum year-class distribution for tautog collected in 2004. Distributions are broken down by sex.

Otoliths — Measurements of reader self-precision were high, with both readers able to reproduce the ages of previously read otoliths (reader 1's CV = 2.9% and reader 2's CV = 1.6%). There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 24.2$, df = 19, $P = 0.18$). In Figure 5 we present a graph of the results for between-reader otolith ageing precision. The average between-reader coefficient of variation (CV) of 1.32 % was not significant.

Of the 501 fish aged with otoliths, 16 age-classes (2 through 16, and 20) were represented. The average age for the sample was 4.7 years. The standard deviation and standard error were 2.7 and 0.1, respectively.

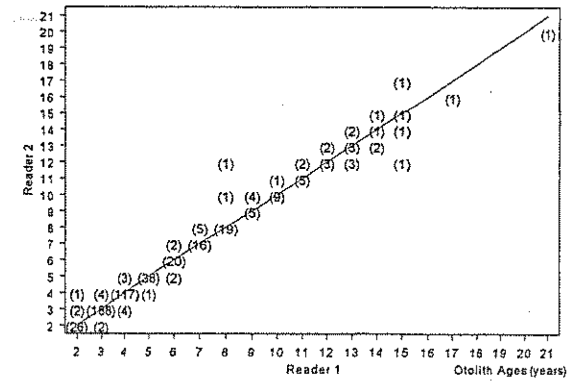


Figure 5. Between-reader comparison of otolith age estimates for tautog.

Comparison of Operculum and Otolith Ages — The between-structure average CV of 6.8% was comparable to the within structure CV's. There was evidence of systematic disagreement between otolith and operculum ages (test of symmetry, $\chi^2 = 66.8$, df = 33, $P < 0.01$). Operculum were assigned a lower age than otoliths for 9% of the fish and 26% of the time were operculum assigned a higher age than otoliths (Figure 6). There was also evidence of bias between otolith and scale ages using an age bias plot (Figure 7), again with

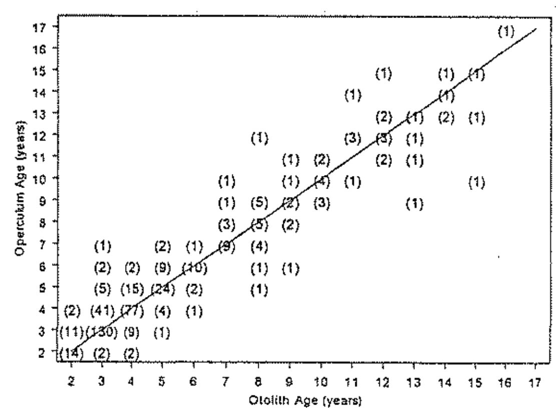


Figure 6. Comparison of otolith and operculum age estimates for tautog.

operculum generally assigned higher ages for younger fish and lower ages for older fish than otoliths age estimates.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using operculum ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

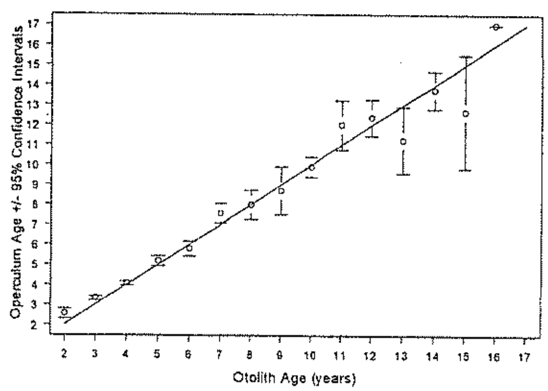


Figure 7. Age-bias plot for tautog otolith and operculum age estimates.

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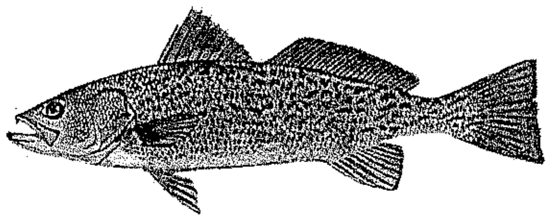
Table 1. The number of tautog assigned to each total length-at-age category for 506 fish sampled for operculum age determination in Virginia during 2004 (operculum not collected for 62 fish and length not reported for 7 fish).

Length 1-inch intervals	Age (years) 2	3	4	5	6	7	8	9	10	11	12	13	14	15	17	Totals
10 - 10.99	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1
12 - 12.99	2	1	0	0	0	0	0	0	0	0	0	0	0	0	0	3
13 - 13.99	3	34	12	0	0	0	0	0	0	0	0	0	0	0	0	49
14 - 14.99	9	59	31	8	1	1	0	0	0	0	0	0	0	0	0	109
15 - 15.99	2	38	30	7	6	1	2	1	0	1	0	0	0	0	0	88
16 - 16.99	1	15	25	11	5	2	3	2	2	1	0	0	0	0	0	67
17 - 17.99	1	4	16	12	7	0	4	1	2	0	0	0	0	0	0	47
18 - 18.99	0	1	7	5	4	3	1	2	2	0	2	1	1	1	0	30
19 - 19.99	0	0	4	0	1	1	0	1	0	1	1	1	0	1	1	12
20 - 20.99	0	0	0	1	1	8	0	3	1	2	3	2	1	0	0	22
21 - 21.99	0	0	0	0	0	1	0	0	0	1	1	1	0	0	0	4
22 - 22.99	0	0	0	0	0	0	0	0	1	0	1	1	0	0	0	3
23 - 23.99	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	1
30 - 30.99	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	1
Totals	18	152	126	44	25	17	10	11	8	6	8	6	2	3		437

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-class, based on operculum ages for tautog sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years) 2	3	4	5	6	7	8	9	10	11	12	13	14	15	17	N
10 - 10.99	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1
12 - 12.99	0.667	0.333	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	3
13 - 13.99	0.061	0.694	0.245	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	49
14 - 14.99	0.083	0.541	0.284	0.073	0.009	0.009	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	109
15 - 15.99	0.023	0.432	0.341	0.080	0.068	0.011	0.023	0.011	0.000	0.011	0.000	0.000	0.000	0.000	0.000	88
16 - 16.99	0.015	0.224	0.373	0.164	0.075	0.030	0.045	0.030	0.030	0.015	0.000	0.000	0.000	0.000	0.000	67
17 - 17.99	0.021	0.085	0.340	0.255	0.149	0.000	0.085	0.021	0.043	0.000	0.000	0.000	0.000	0.000	0.000	47
18 - 18.99	0.000	0.033	0.233	0.167	0.133	0.100	0.033	0.067	0.067	0.000	0.067	0.033	0.033	0.033	0.000	30
19 - 19.99	0.000	0.000	0.333	0.000	0.083	0.083	0.000	0.083	0.000	0.083	0.083	0.083	0.000	0.083	0.083	12
20 - 20.99	0.000	0.000	0.000	0.045	0.045	0.364	0.000	0.136	0.045	0.091	0.136	0.091	0.045	0.000	0.000	22
21 - 21.99	0.000	0.000	0.000	0.000	0.000	0.250	0.000	0.000	0.000	0.250	0.250	0.250	0.000	0.000	0.000	4
22 - 22.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.333	0.000	0.333	0.333	0.000	0.000	0.000	3
23 - 23.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1
30 - 30.99	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000	0.000	1
														Sample Size		437

Chapter 13
Weakfish



Cynoscion regalis

INTRODUCTION

A total of 657 weakfish, *Cynoscion regalis*, was collected by the VMRC's Stock Assessment Program for age and growth analysis in 2004. The average age was 2.3 year old, and the standard deviation and standard error were 1.0 and 0.04, respectfully. Nine age classes (1 to 5, and 7 to 10) were represented, comprising fish from the 1994-1997 and 1999-2003 year-classes, with fish primarily from the 2001 through 2003 year-classes.

METHODS

Handling of collection — Otoliths were received by the Age & Growth Laboratory in labeled coin envelopes. Once in our hands, they were sorted based on date of capture, their envelope labels were verified against VMRC's collection data, and assigned unique Age and Growth Laboratory sample numbers. All otoliths were stored dry in labeled cell well trays.

Preparation — The first step in otolith preparation was to grind down the otolith in a transverse plane to its core using a Hillquist thin section machine's 320-mesh diamond cup wheel. To prevent distortion of the reading surface, the otolith was ground exactly perpendicular to the reading plane. The otolith's ground surface was then placed face down in a drop of Loctite 349 photo-active adhesive on a labeled glass slide and placed under ultraviolet light to allow the adhesive to harden. The Hillquist thin section machine's cup wheel was used again to grind the otolith, embedded in Loctite, to a thickness of 0.3 to 0.5 mm. Finally, a thin layer of Flo-texx mounting medium was applied to the otolith section to increase light transmission through the translucent zones, which provided enhanced contrast and greater readability.

Readings — Two different readers aged all sectioned otoliths using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization at between 8 and 100 times magnification (Figure 1). Each reader aged all of the otolith sections using ageing criteria listed in Lowerre-Barbieri et al. (1994). All samples were aged in chronological order based on collection date, without knowledge of previously estimated ages or the specimen lengths. When the readers' ages agreed, that age was assigned to the fish. When the two readers

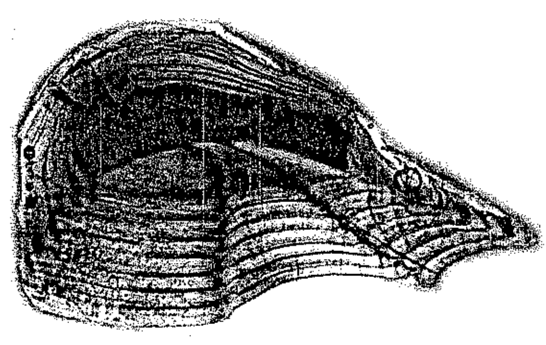


Figure 1. Sectioned otolith from a 7 year old female weakfish.

disagreed, both readers sat down together and re-aged the fish, again without any knowledge of previously estimated ages or lengths, and assigned a final age to the fish. When the readers were unable to agree on a final age, the fish was excluded from further analysis.

Comparison Tests — Age estimates from reader 1 were plotted against age estimates from reader 2 to assess deviation from 1:1 equivalence (Campana et al. 1995). A test for symmetry was used to detect any systematic difference between the two readers (Hoenig et al. 1995). Also, a random sub-sample of 50 fish was selected for second readings to measure reader precision and age reproducibility. We considered a reader to be biased if the readings revealed consistent over or under ageing.

RESULTS

The measurement of reader self-precision was high for both readers (reader 1's CV = 0% and reader 2's CV = 0%). There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 9$, df $= 6$, $P = 0.17$). Figure 2 illustrates the between readers' precision of age estimates. The average coefficient of variation (CV) of 0.5% was not significant.

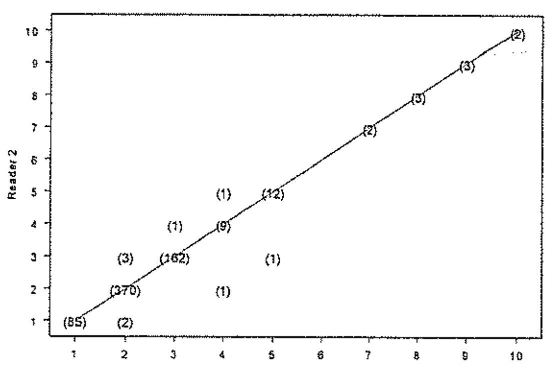


Figure 2. Between-reader comparison of otolith age estimates for weakfish.

Of the 657 fish aged with otoliths, nine age classes were represented (Table 1). The average age was 2.3 years old, and the standard deviation and standard error were 1.0 and 0.04, respectfully.

Year-class data (Figure 3) show that the fishery was comprised of nine year-classes, comprising fish from the 1994-1997 and 1999-2002 year-classes, with fish primarily from the 2001 through 2003 year-classes.

Age-Length-Key — In Table 2 we present an age-length-key that can be used in the conversion of numbers-at-length in the estimated catch to numbers-at-age using otolith ages. The table is based on VMRC's stratified sampling of landings by total length inch intervals.

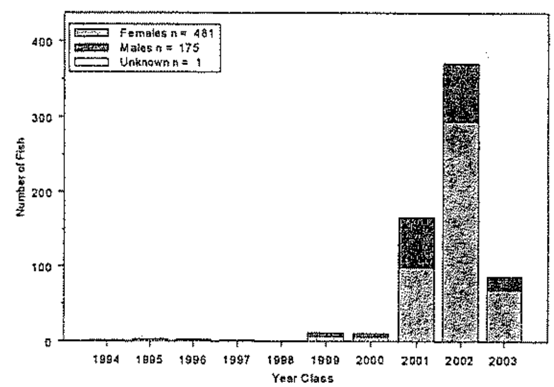


Figure 3. Year-class frequency distribution for weakfish collected for ageing in 2004. Distribution is broken down by sex.

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Table 1. The number of weakfish assigned to each total length-at-age category for 657 fish sampled for age determination in Virginia during 2004 (no lengths for 2 fish)

Length 1-inch intervals	Age (years)									
	1	2	3	4	5	7	8	9	10	Totals
7 - 7.99	1	0	0	0	0	0	0	0	0	1
8 - 8.99	4	0	0	0	0	0	0	0	0	4
9 - 9.99	33	27	1	0	0	0	0	0	0	61
10 - 10.99	26	114	8	0	0	0	0	0	0	148
11 - 11.99	8	146	45	1	0	0	0	0	0	200
12 - 12.99	10	62	50	2	0	0	0	0	0	124
13 - 13.99	1	14	25	1	0	0	0	0	0	41
14 - 14.99	3	1	12	1	1	0	0	0	0	18
15 - 15.99	1	1	8	1	1	0	0	0	0	12
16 - 16.99	0	3	6	1	0	0	0	0	0	10
17 - 17.99	0	2	1	0	0	0	0	0	0	3
18 - 18.99	0	0	4	0	0	0	0	0	0	4
19 - 19.99	0	0	1	2	0	0	0	0	0	3
20 - 20.99	0	0	2	1	1	0	0	0	0	4
22 - 22.99	0	0	0	1	0	0	0	0	0	1
23 - 23.99	0	0	3	0	0	0	0	0	0	3
27 - 27.99	0	0	0	0	3	0	0	0	0	3
28 - 28.99	0	0	0	0	4	0	0	0	0	4
29 - 29.99	0	0	0	0	2	1	1	1	0	5
30 - 30.99	0	0	0	0	0	1	1	0	0	2
31 - 31.99	0	0	0	0	0	0	1	2	1	4
34 - 34.99	0	0	0	0	0	0	0	0	1	1
Totals	87	370	166	11	12	2	3	3	2	655

Table 2. Age-Length key, as proportions-at-age in each 1 inch length-intervals, based on otolith ages for weakfish sampled for age determination in Virginia during 2004.

Length 1-inch intervals	Age (years) 1	2	3	4	5	7	8	9	10	N
7 - 7.99	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1
8 - 8.99	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	4
9 - 9.99	0.541	0.443	0.016	0.000	0.000	0.000	0.000	0.000	0.000	61
10 - 10.99	0.176	0.770	0.054	0.000	0.000	0.000	0.000	0.000	0.000	148
11 - 11.99	0.040	0.730	0.225	0.005	0.000	0.000	0.000	0.000	0.000	200
12 - 12.99	0.081	0.500	0.403	0.016	0.000	0.000	0.000	0.000	0.000	124
13 - 13.99	0.024	0.341	0.610	0.024	0.000	0.000	0.000	0.000	0.000	41
14 - 14.99	0.167	0.056	0.667	0.056	0.056	0.000	0.000	0.000	0.000	18
15 - 15.99	0.083	0.083	0.667	0.083	0.083	0.000	0.000	0.000	0.000	12
16 - 16.99	0.000	0.300	0.600	0.100	0.000	0.000	0.000	0.000	0.000	10
17 - 17.99	0.000	0.667	0.333	0.000	0.000	0.000	0.000	0.000	0.000	3
18 - 18.99	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	4
19 - 19.99	0.000	0.000	0.333	0.667	0.000	0.000	0.000	0.000	0.000	3
20 - 20.99	0.000	0.000	0.500	0.250	0.250	0.000	0.000	0.000	0.000	4
22 - 22.99	0.000	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	1
23 - 23.99	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	3
27 - 27.99	0.000	0.000	0.000	0.000	1.000	0.000	0.000	0.000	0.000	3
28 - 28.99	0.000	0.000	0.000	0.000	1.000	0.000	0.000	0.000	0.000	4
29 - 29.99	0.000	0.000	0.000	0.000	0.400	0.200	0.200	0.200	0.000	5
30 - 30.99	0.000	0.000	0.000	0.000	0.000	0.500	0.500	0.000	0.000	2
31 - 31.99	0.000	0.000	0.000	0.000	0.000	0.000	0.250	0.500	0.250	4
								Sample Size		655

Chapter 14 Validation of otolith-based ageing and a comparison of otolith-and opercula-based ageing of tautog *Tautoga onitis.*

Introduction

Tautog (*Tautoga onitis*) is a recreationally and commercially-targeted fish that ranges in coastal waters from Nova Scotia (Bleakney, 1963; Bigelow and Schroeder, 1953) to Georgia (Parker, 1990). Catches from the recreational and commercial fishery were highest in 1987 with a peak catch of 250 metric tons (Personal communication from the National Marine Fisheries Service, Fisheries Statistics Division 2004). While juvenile tautog move relatively little and have high site fidelity (Able et al. 2005), adults migrate each fall from inshore summering grounds to offshore overwintering sites, triggered by temperature (Olla et al. 1980), and spawn from April to June in the lower Chesapeake Bay and coastal waters of Virginia (White et al. 2003). Adult tautog are generally found inhabiting manmade or natural complexly structured habitats (Steimle, 1999), feeding primarily on mussels and various crustaceans (Bigelow and Schroeder, 1953; Olla et al, 1974). It is currently believed that due to slow growth rate, late age at maturity, and predictable habitat residence that this species is subject to overexploitation (Hostetter and Munroe, 1993). Although the primary literature contains information on basic biology and ecology of this species (Arendt et al, 2001; Bleakney, 1963; Cooper, 1966; Olla et al, 1974; Olla and Samet, 1977; Parker, 1990; White et al 2003), very few studies describe age and growth of the species (Cooper, 1967; Hostetter and Munroe, 1993) and these are based on opercular ageing.

Fisheries management of tautog is based on age estimates derived from their opercula. The method of reading tautog opercula was developed by Cooper (1967), who estimated a maximum age of 34 years for males and 22 years for females. Subsequently, other scientists have continued to use this method. For example, Simpson (1989) estimated a maximum of age 31 for males and 25 for females. For the Chesapeake Bay region, Hostetter and Munroe (1993) reported that both male and female tautog matured at age 3, and the maximum ages were 25 and 21, respectively. Typically opercula ages result in lower precision compared to other structures (Sipe and Chittenden, 2001), but when Hostetter and Munroe (1993) compared tautog opercula and otoliths, they found that opercula were more precise and therefore the hard part of choice. However, they evaluated opercula to whole otoliths and their sample size was limited to a small number of age classes. Theirs was an inadequate test because whole otoliths have been found to be less precise and can underage fish compared to sectioned otoliths (Beamish and McFarlane, 1983). A better comparison would be between opercula and sectioned otoliths. Moreover, new procedures have made sectioning practical for even the smallest and most fragile of otoliths, making tautog otoliths simple to process, similar to that done for tautog's closest relative: cunner (*Tautogolabrus* adspersus) (Nitschke and Burnett, 2001).

Before the use of sectioned otoliths can replace opercula for routine ageing of tautog, we must: 1) validate annuli formation in otoliths, 2) demonstrate that precision can be sufficiently improved so that the additional preparation effort to use otoliths is worthwhile, and 3) subsequent use of otolith ages provide comparable estimates of growth as that derived from opercula such that there is continuity in stock assessments. In a side-by-side comparison between sectioned otoliths and opercula, we will use marginal increment analysis (MIA) to validate annuli formation in otoliths, and measure within and between reader precision. Marginal increment analysis has already been used to validate annulus formation on tautog opercula (Simpson 1989; Hostetter and Munroe 1993), but note that opercula are troublesome when trying to age young fish because the basal region is obscure and difficult to read, with the potential result that the first few years are misread.

Comparisons have shown that sectioned otoliths provide accurate and precise age estimates for a variety of fish species. For example, Erickson (1983) compared walleye (*Stizostedion vitreum*) age estimates using otoliths, dorsal spines, and scales, and found that sectioned otoliths provided more accurate age estimates. Barber and McFarlane (1987) evaluated ageing techniques using whole otoliths, burnt sectioned otoliths, and sectioned fin rays, and concluded that burnt sectioned otoliths gave the best age estimates for Arctic char (*Salvelinus alpinus*). Lowerre-Barbieri (1994) found that weekfish (*Cynoscion regalis*) age estimates are more precise using otoliths than using scales and other hard parts. Welch et al. (1993) and Secor et al. (1995) found that sectioned otoliths yielded more accurate age estimates especially for older striped bass (*Morone saxatilis*) than did scales. To date, no studies have compared opercula and sectioned otoliths for ageing tautog, although opercula have been used to age this species for almost 40 years. However, such information is crucial to better understanding tautog life history and for improving management of this species in the mid-Atlantic region.

Methods

Data collection

Tautog were collected from the commercial fishery in Chesapeake Bay between 2000 to 2004 from the following gears: hand-lines, otter trawls, gill nets, pound nets, and pots. A total of 1913 fish were collected from all months of the year. Upon collection, each specimen was measured for standard length (SL, $\pm$1.0mm), total length (TL, $\pm$1.0mm) and total weight (TW $\pm$1.0g). Sexual determination was made using macroscopic gonad analyses. Both sagittal otoliths were extracted and stored in vials. Because Cooper (1967) found no major differences between the ages of right and left opercula, we removed only right-side opercula for ageing. The opercula were put in coin envelopes and stored in a refrigerator immediately. All the hard parts were labeled and kept in date-of-capture chronological order for further processing.

Hard part processing

Otoliths

Left or right otoliths were randomly chosen for processing. The processing consists of three steps as follows:

1. Baking - Each otolith was placed into a ceramic "Coors" spot plate well and baked in a Thermolyne 1400 furnace at 400°C. Baking time was dependent on otolith size and gauged to achieve a light caramel color.

2. Embedding - The baked otolith was embedded in a mold that was half filled with Loctite 349 adhesive, by placing the otolith parallel to the long axis of the mold. When dry, the mold was filled completely with more Loctite and when dry the position of the otolith core was marked using a fine point felt pen.

3. Sectioning - A 0.5 mm thin transverse section which encompassed the marked core was made using a low speed Buelher Isomet saw equipped with two Norton diamond wafering blades and a 0.5 mm spacer. The thin section was then adhered to a labeled glass slide using Crystal Bond™. If necessary, the section was polished on a Buehler Ecomet 3 variable speed grinder-polisher with Mark V Laboratory 30-micron polishing film. To enhance contrast and insure greater readability, the thin section was covered with a thin layer of Flo-texx mounting medium.

Opercula

Tautog operculum bones were boiled in water for several minutes to remove excess tissue. Opercula were then air dried and transferred to labeled coin envelopes for dry storage in preparation for ageing.

Otolith annulus validation

We used marginal increment analysis (MIA) to validate otolith ageing (Campana 2001). The marginal increment was defined as the distance from the beginning of the last dark narrow band to the edge of the otolith. The marginal increment was measured to the nearest 0.001 mm along the ventral side of the sulcal groove using a digital image system (Image-Pro Plus 5.0 MediaCybernetics, 5x magnification, a Hitachi KP-D50 color digital camera, and a Leica MZ12 microscope). Otolith samples were arranged first randomly by year and then chronologically by capture date within the year to prevent any bias with the measurements (Campana 2001). An average of the marginal increments for each month was calculated using the data of all years and ages combined.

Ageing

Opercula and otoliths were aged separately to avoid ageing bias. We assigned ages based on annulus counts using a January 1 birthday. Two different readers separately aged both calcified structures. Both readers aged all samples in chronological order based on collection date, without knowledge of specimen length or previously estimated ages of either hard-part. When the reader's ages agreed, then that age was assigned to the fish. When they disagreed, both readers re-aged the fish together without any knowledge of previously estimated ages, and assigned a final age to the fish. If the readers were still unable to reach agreement, the fish was excluded from further analysis. To measure reader self-precision and age reproducibility, each reader re-aged a randomly selected subset of 50 fish from each year of sampling.

Otoliths

Otolith sections were aged using a Leica MZ-12 dissecting microscope with transmitted light and dark-field polarization between 8 and 100 times magnification. An annual growth band was defined as the combination of a dark caramel band and a very light caramel band, and the dark caramel band was defined as an annulus. The otolith core was a dark caramel zone at the ventral to the posterior side, just above the sulcal groove (Figure 1). Dark caramel bands were counted for ageing and the reading plane was the ventral side of the sulcal groove beginning at the core and continuing distally to the ventral edge. The first annulus was the first distinct dark caramel band subsequent to the core.

Opercula

Cooper (1967) defined an annual growth band as a combination of an opaque and a translucent band on an operculum, and a sharp transition from translucent to opaque as an annulus (Figure 2). Our descriptions of an annual growth band and an annulus are similar to Coopers' with a slight modification. Our observations indicate that opaque and translucent bands were not divided as clearly as Cooper's definition. We found that an annual growth band starts with a dark narrow opaque band that gradually became lighter and lighter, and eventually transitioned to a relatively wide translucent band. The next annual growth band repeats this pattern. Therefore, we have defined an annulus as a sharp transition from the translucent band to the beginning of the dark narrow opaque band. In addition, we observed some visible lines occurred within such patterns and defined them as false annuli or checks. Also, an incomplete dark narrow opaque band (one that did not cross from one side to the other of operculum) was considered as a check.

Hostetter and Munroe (1993) found that the entire first annulus was often not clearly visible due to thickening of the buttress zone at the articular apex, especially on the opercula of older fish. Therefore, whenever a translucent part appeared right after the buttress zone, we counted one annulus even though we didn't see a dark narrow opaque band (or a complete pattern). Opercula were aged using a light table with no magnification.

Data analysis

Precision

We used a test for symmetry to detect systematic difference between the two readers and within each reader (Hoenig et al., 1995) for both structures. Precision between two readers and within a reader were measured using Chang's (1982) average coefficient of variation (CV) with the formula presented in Campana and Jones (1992).

$$CV_j = 100\% * \frac{\sqrt{\sum_{i=1}^{R} \frac{(X_{ij} - \overline{X}_j)^2}{R - 1}}}{\overline{X}_j} \qquad 1)$$

Where X_{ij} is the ith age determination of the jth fish, X_j is the mean age of the jth fish, and R is the number of times each fish is aged.

$$\text{Mean } CV = \frac{1}{n} \sum_{j=1}^{n} CV_j \quad j = 1, 2, 3, \ldots n, \qquad 2)$$

where n is the total number of fish aged.

Growth Comparisons

Because we lacked one-year old fish in our samples, we back-calculated total length at age 1 from a subset of age 2 fish and excluded this subset from further analysis. The back-calculation equation is as follows:

$$L_1 = \frac{L_c \times O_1}{O_c} \qquad 3)$$

where L_c stands for the measurements of age 2 fish total length at the time of capture. O_c is the otolith radius at the time of capture, and L_1 and O_1 are the corresponding estimate and measurement at the time of formation of the first annulus. We also combined all fish from age 13 to age 24 into an age 13+ group because few fish were in these older age groups.

We fit a von Bertalanffy growth model (Ricker, 1975) to mean total length at age by sex using non-linear least squares regression (SAS Institute 1996):

$$L_t = L_{\inf}(1 - e^{-k(t - t_0)}) \qquad 4)$$

where L_t is observed length at any given age; L_{inf} is asymptotic mean length (mm); t_o is the theoretical age at 0 length; k is the instantaneous growth rate (Brody coefficient) and t is the estimated age.

Next, we examined the differences between between sexes using likelihood ratio tests (Kimura, 1980). Models were developed to assess three hypotheses as follows used:

1. H_0: different growth curves with no parameters equal

$$H_0 : l_{ij} = l_{\infty i}\left(1 - e^{-k_i(t_{ij} - t_{0i})}\right) \qquad 5)$$

2. $H_{1\text{-}3}$: different growth curves with one parameter (L_{inf}, k, or t_0,) equal

$$H_1 : l_{ij} = l_{\infty}\left(1 - e^{-k_i\left(t_{ij} - t_{01}\right)}\right) \quad 6)$$
$$H_2 : l_{ij} = l_{\infty i}\left(1 - e^{-k\left(t_{ij} - t_{01}\right)}\right) \quad 7)$$
$$H_3 : l_{ij} = l_{\infty i}\left(1 - e^{-k_i\left(t_{ij} - t_{0}\right)}\right) \quad 8)$$

3. H_4: all growth curve parameters equal

$$H_4 : l_{ij} = l_{\infty}\left(1 - e^{-k\left(t_{ij} - t_{0}\right)}\right) \quad 9)$$

When comparing two curves using likelihood ratios, an unconstrained model (H_0) is used to compare with more specific models (H_i: $i = 1 \ldots 4$). The statistics are developed by comparing the ratio of the two likelihoods (Kimura, 1980)

$$\Lambda = \frac{\left(2\pi\hat{\sigma}_i^2\right)^{-\frac{N}{2}} e^{\left(\frac{-N}{2}\right)}}{\left(2\pi\hat{\sigma}_0^2\right)^{-\frac{N}{2}} e^{\left(\frac{-N}{2}\right)}} = \left(\frac{\hat{\sigma}_0^2}{\hat{\sigma}_i^2}\right)^{\frac{N}{2}} \quad 10)$$

with the test statistic:

$$-2Ln(\Lambda) = -NLn\left(\frac{\hat{\sigma}_0^2}{\hat{\sigma}_i^2}\right) \quad 11)$$

which takes a chi-squared distribution and is used to test the significance of differences between the unconstrained and specifically-constrained models (Kimura, 1980).

Results

Of the 1913 tautog collected from 2000 to 2005, 1860 and 1814 fish were aged using otoliths and opercula, respectively, making a total of 1908 fish aged (Table 1). Otolith ages ranged from 2 to 25 with a mean age of 5.11 overall. Opercula ages ranged from 0 to 20 with a mean age of 5.13 overall.

Marginal increment analysis

Marginal increment analysis revealed minimal growth from the last annulus to the edge during the months of July and August for all age classes (Figure 3). This graphical illustration validates that only one opaque zone is deposited on an otolith each year and demonstrates that sectioned otoliths can be used reliably for ageing

Precision

Precision for otolith-based ages

The average coefficients of variation (CV) within Reader 1 and within Reader 2 were 1.2% and 1.0%, respectively (Table 2). The average CV between two readers was 1.3%. There was no evidence of systematic disagreement between Reader 1 and Reader 2 (test of symmetry, $\chi^2 = 27.3$, df = 18, P = 0.09).

Precision for operculum-based ages

Precision both within reader and between readers was lower than corresponding precision for otolith-based ages (Table 2). The average CV within Reader 1 and within Reader 2 were 3.7% and 5.1%, respectively. The average CV between two readers was 6.8%. Ageing precision for older fish ($\geq$ age 5, n = 830) was similar to younger fish. There was no evidence of systematic disagreement between reader 1 and reader 2 (test of symmetry, $\chi^2 = 21.4$, df = 15, P = 0.12).

Comparisons of growth models

Likelihood ratio tests indicated that male and female growth rates were significant different in our study (Table 3). Growth rates for both males and females in this study were significantly different from those in the study of Hostetter and Munroe (1993). We found that tautog grew faster with smaller average maximum total length compared to their study (1993). The greatest incremental growth in TL occurred during the first and second year for both males and females (Figure 4).

Discussion

This study is the first to evaluate the use of sectioned otoliths for ageing tautog. A vast literature exists to show that sectioned otoliths are almost always the preferred hard part for ageing because they provide ages more accurately and precisely. Even though Hostetter and Munroe (1993) compared opercular and otolith ageing and found opercula were more precise, they used only the whole otoliths, which are not as reliable as sectioned otoliths. Using only 27 of 712 tautog, they concluded that opercula provided more accurate age estimates especially for older tautog. This study overcame the shortcomings in their study by examining sectioned otoliths collected from nearly 2000 fish sampled over a 5-year period. Not surprisingly with larger sample size and upon sectioning, otoliths prove to be both accurate and more precise for ageing tautog. Thus, we conclude that it is appropriate to use otolith thin sections for ageing.

Even though otolith sections take more time to prepare, they may be better overall than opercula for several important reasons. Although they take longer to prepare, they are easier to read and reading them takes less time. When a study demands large numbers to age, then the overall time to age with sectioned otoliths may be the same or less than with opercula. Otolith sections lack the problem of an obscure basal region that is found in opercula, especially for older fish. Finally, otoliths exhibited virtually no checks or false annuli, a problem that occurred in opercula.

When we used otolith age to model growth, the growth rate for young tautog of 3 years or less are dramatically faster than that found in previous studies which were based on opercula. This could be due to temporal differences in growth, which we cannot dismiss, but is most likely due to ageing errors in opercula. Our modified method of opercula ageing is specifically designed for identifying checks on the opercula of younger tautog and this may have been one of the problems with earlier work. There is some indication in previous studies on other fish species indicate that spawning and migration induced by temperature can produce checks on calcified structures (Summerfelt and Hall 1987). Olla et al. (1980) reported that adult tautog had both spawning and migration resulted from temperature increases in April and September. We also observed that such checks appeared predominantly among the first few annuli on tautog opercula. Therefore, it is not unreasonable to believe that previous opercula ageing method may have counted spawning and migration checks as annuli, thus overestimating age when tautog especially in younger fish. This may also be the reason that the maximum age of tautog found in this study is younger than those found in previous studies. For example, Cooper (1967) did not report whether he found any checks on tautog opercula and how to distinguish them from annuli.

The increased growth rates in younger tautog and decreased maximum age found in this study could influence the management of tautog dramatically. Tautog have long been considered as slow-growing and long-lived species (Hostetter and Munroe 1993). Combining this traditional knowledge with other biotic and abiotic information, ASFMC in 1993 has declared that the tautog population in Atlantic region has been overexploited. Our study suggests that tautog are younger based on otolith ageing and that stock assessment scientists may need to revise their estimates given our new data. However, the current management strategy will protect the stocks because they result in more conservative policy decisions.

In summary, this study used otolith ageing as a reference to evaluate opercula ageing, an approach that has proved to be effective in many previous studies on other hard parts such as scales, opercula, and fin rays, etc (Erickson 1983; Barber and McFarlane 1987; Welch et al. 1993; Lowerre-Barbieri 1994; Secor et al. 1995). Considering our results could influence tautog fishery management dramatically if applied, we suggest that future studies should focus on identifying spawning and migration checks on young tautog opercula using a variety of direct evaluation techniques such as oxytetracycline marking, if possible. Such verification combined with the results from this study will improve our understanding of tautog population dynamics and enhance its fishery management in the mid-Atlantic region.

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Table 1

A summary of the number of tautog (*Tautoga onits*) by month and sex used for age estimates and marginal increment analysis (MIA).

	Ageing				MIA			
Month	Male	Female	Unknown	Totals	Male	Female	Unknown	Totals
January	68	48	0	116	62	45	0	107
February	27	33	0	60	24	26	0	50
March	110	105	3	218	97	78	3	178
April	44	51	3	98	41	44	3	88
May	128	153	1	282	122	136	1	259
June	74	61	5	140	61	53	5	119
July	10	9	0	19	10	5	0	15
August	21	12	0	33	19	9	0	28
September	6	12	0	18	4	7	0	11
October	202	161	1	364	193	149	1	343
November	271	210	2	483	259	188	2	449
December	42	32	3	77	37	28	3	68
Totals	1003	887	18	1908	929	768	18	1715

Table 2

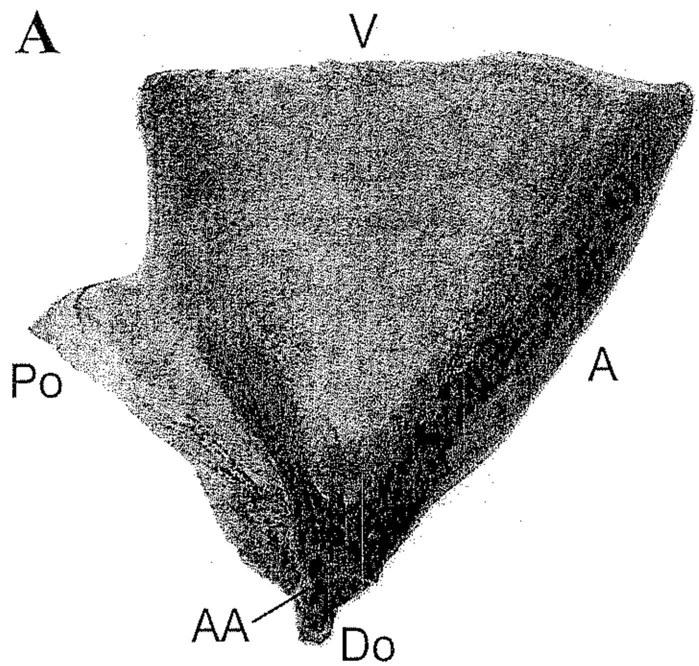
Overall percent agreement and percent agreement (±1 band) for both sexes combined on presumed annual marks on tautog (*Tautoga onitis*) otoliths and opercular bones.

Structure	Precision Reader 1/2	Coefficient of Variation Reader 1/2	Between Reader	Between Reader ±1Band
Operculum	79.4/66.0	3.7/5.11	61.8	92.2
Otolith	92.0/91.6	1.0/0.92	89.7	98.9

Table 3. Likelihood ratio tests comparing von Bertalanffy parameter estimates for males and females in this study, and for males and females between Hostetter and Munroe (1993)'s study and this study. N is the number of age groups in each comparison (sample size).

Hypothesis	Linear Constraints	l_1	l_2	k_1	k_2	t_1	t_2	RSS	Chi-square	df	P
		Female and Male in This Study (Otoliths), N = 26, where subscript 1=female, 2=male									
H0	None	520.2	549.5	0.2403	0.2767	-1.7285	-1.1434	11460			
H1	The same l	542.4	542.4	0.1931	0.2975	-2.3458	-0.9948	11792	0.74	1	0.3888
H2	the same k	513.7	554.1	0.2616	0.2616	-1.4956	-1.2818	11533	0.17	1	0.6845
H3	the same t	512.2	555.5	0.2698	0.2549	-1.3781	-1.3781	11624	0.37	1	0.5433
H4	the same l, k, and t	534.2	534.2	0.2604	0.2604	-1.3949	-1.3949	17496	11.00	3	0.0117
		Males between Hostetter and Munroe (1993)'s Study (Opercula) and This Study (Otoliths), N = 36, where subscript 1 = Hostetter and Munroe (1993), 2 = this study									
H0	None	718.1	549.5	0.0884	0.2767	-3.2355	-1.1434	18392			
H1	the same l	696.8	696.8	0.0975	0.1008	-2.8738	-4.2654	23928	9.47	1	0.0021
H2	the same k	695.5	690.9	0.0994	0.0994	-2.7481	-4.5238	23965	9.52	1	0.002
H3	the same t	697.7	590.2	0.0997	0.1758	-2.6126	-2.6126	20548	3.99	1	0.0458
H4	the same l, k, and t	682	682	0.1036	0.1036	-3.4765	-3.4765	37803	25.94	3	<0.0001
		Females between Hostetter's Study (Opercula) and This Study (Otoliths), N = 33, where subscript 1 = Hostetter and Munroe (1993), 2 = this study									
H0	None	691.3	520.2	0.0877	0.2403	-3.3816	-1.7285	24482			
H1	the same l	667.5	667.5	0.0976	0.0881	-3.0034	-5.2449	27612	3.97	1	0.0463
H2	the same k	665.4	635.3	0.0996	0.0996	-2.8905	-4.899	27160	3.43	1	0.0641
H3	the same t	672	550.2	0.0972	0.1716	-2.9075	-2.9075	25229	0.99	1	0.3193
H4	the same l, k, and t	534.2	534.2	0.2604	0.2604	-1.3949	-1.3949	35299	12.08	3	0.0071

Figure 1.
(**A**) Photograph of a *T. onitis* right whole opercular bone depicting orientation (**V**) ventral; (**Do**) dorsal; (**Po**) posterior; (**A**) anterior. Total length from dorsal to ventral edge is 45mm. (**B**) Photograph showing *Tautoga onitis* otolith thin section. Orientation of otolith is (**P**) proximal; (**Di**) distal; (**Do**) dorsal; (**V**) ventral: (**SG**) sulcal groove. Black arrow illustrates measurement used for MIA analysis. Total length from dorsal to ventral edge is 1.809mm.



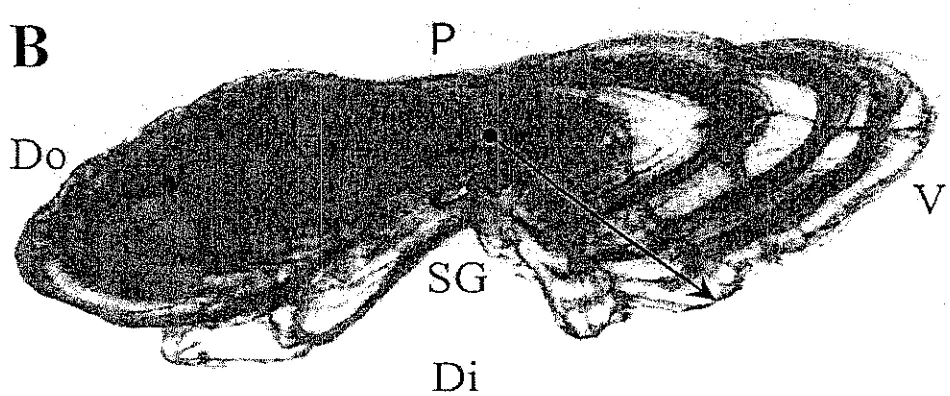
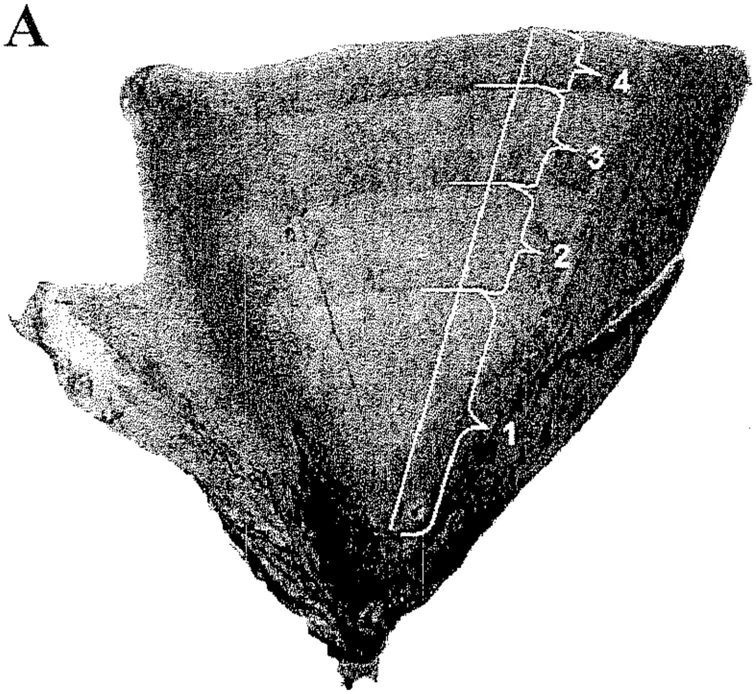


Figure 2.
Photograph illustrating *Tautoga onitis* annuli from two different hard parts. (**A**) Right whole opercular bone (TL 47.0mm) of a four-year-old fish. Vertical white line indicates reading plane. The area between two horizontal white lines represents one year's growth. (**B**) *T. onitis* otolith (TL 2.246mm) of a six-year-old fish. Black arrows and numbers indicate annuli and white oval denotes core.

A



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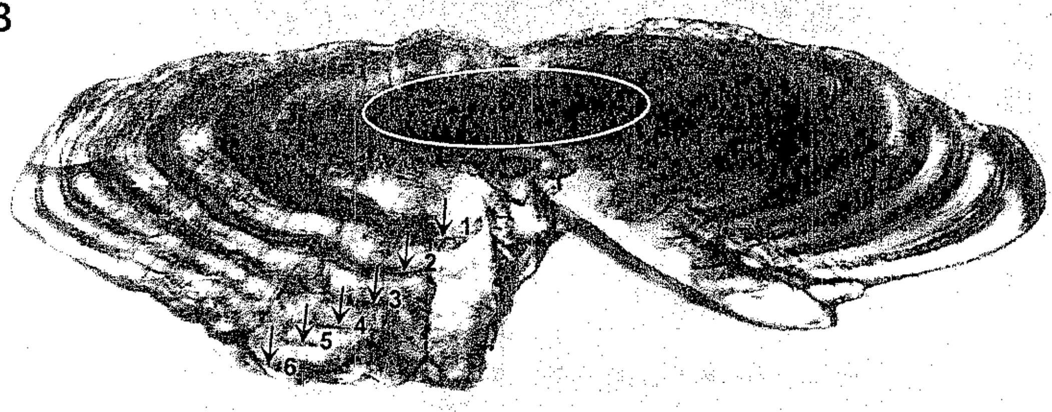

Figure 3: Mean seasonal incremental growth (in mm) of Tautog otoliths from Chesapeake Bay and Virginia coast water in 2000-2004. Vertical bars represent the 95% confidence intervals and n is the total sample size.

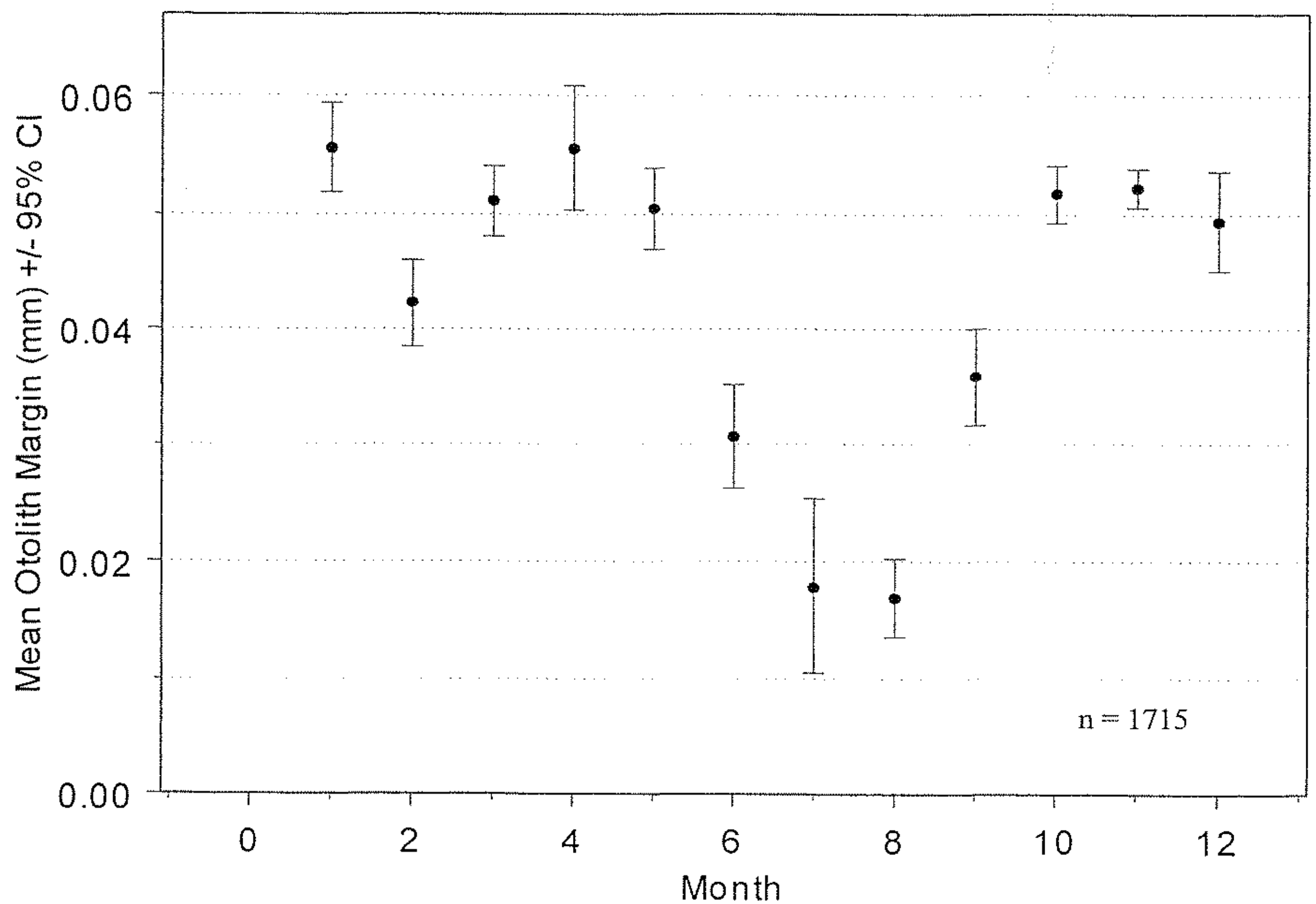

Figure 4. Estimated mean total lengths at age and von-Bertalanffy growths for both males and females in Chesapeake Bay and coastal waters of Virginia, using the otolith-based method in this study. The data are year-pooled from 2000 to 2004. The solid square and empty circle represent estimated mean total lengths for both males and females, respectively. The dash line and solid line represent von-Bertalanffy growths for both males and females, respectively.